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NEW-PLACE EFFECT IN MAIZE

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INTRODUCTION

Widely divergent opinions have been expressed regarding the advisability of transferring seed from one region to another. With maize (*Zea mays*) the transfer of seed is generally held to be disadvantageous. Numerous experiments have shown that when seed of the same original variety, but grown at two places, is planted side by side at one of the places the results are in favor of the local seed.

The relative superiority of the locally selected seed has been so pronounced that the securing of seed from distant localities, except where grown for forage, has been discouraged. The natural result is to confine the utilization of carefully selected strains to the localities where the breeding is done. Caution in transferring seed is certainly desirable, but the results of the experiments here reported indicate that two opposing factors are involved, the relative importance of which must be determined before generalizations are made.

The differences shown between the geographically separated lines of the same variety when brought together in the locality where one of the lines has been grown may be ascribed to three general causes:

(1) Cross-pollination. Varieties removed to different localities may become crossed with other sorts, with the result that their characteristics are changed in a greater or less degree.

(2) Different standards of selections. Changed conditions in connection with the diversity that exists in all varieties bring slightly different types of plants into prominence, and selection, either conscious or unconscious, results in a changed type.

(3) New-place effects. The more or less temporary changes that follow a transfer to new conditions, caused by the novelty or "shock" of the new environment without special reference to the nature of the change.

So far as known, new-place effect, which the experiments here reported show to be a significant factor in the transfer of seed, has not previously been considered with regard to maize.

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These experiments were originally planned with the idea that the seed of a first-generation hybrid might be transferred to a distant locality without showing the reduced yields thought to follow the transfer of pure strains.

It was believed that with first-generation hybrid seed it might be a matter of indifference where the hybrid was made, provided the parents were suited to the localities where the crop was to be grown.[1] If found to be the case, this would constitute an important addition to the advantages that follow the use of first-generation hybrid seed, making possible a wider application of the results secured through the work of skilled breeders.

The results indicate that hybrids made in one locality and grown in another are not only at no disadvantage compared with the same hybrids produced at the locality where the comparison is made but that the introduced hybrid may even be superior. It further appears that the same is true of pure strains and that, after the effects of cross-pollination and selection have been eliminated from the problem, there is a residual effect of the transfer to the new place that tends to increase rather than to reduce the vigor and yield of the plants.

NATURE OF THE EXPERIMENTS

It was planned to conduct the experiments at the following places: Stockton, Kans.; Victoria, Tex.; and Lanham, Md. These places represent a wide range of soil and climate. At Stockton, which is in the eastern part of the semiarid Great Plains area, there is a fertile friable soil, low rainfall, low atmospheric humidity, and a prevalence of high winds. Victoria, located in the Gulf region of Texas, has a stiff black clay soil of good fertility. The total rainfall is usually large and the humidity high, though severe drouths are not uncommon. At Lanham, a few miles north of Washington, D. C., the soil is sandy, acid, and relatively infertile. The rainfall and humidity are usually neither deficient nor excessive. In the discussion of results these localities will be referred to as Kansas, Texas, and Maryland.

Four varieties of maize were chosen for the experiment, as follows:

(1) Stockton, a white dent variety developed at Stockton, Kans.

(2) Strawberry, a large-eared Texas variety with red and white variegated dent seed, well adapted to the conditions at Victoria, Tex.

(3) Hickory King, a strain of this variety grown in Virginia and well suited to the conditions at Lanham, Md.

(4) Boone, a strain of "Boone County White," the seed of which was obtained from Illinois.

In the spring of 1912 seed of the four varieties were planted at each of the three places named. The precaution was taken to mix the seed

[1] COLLINS, G. N. THE VALUE OF FIRST-GENERATION HYBRIDS IN CORN. U. S. Dept. Agr. Bur. Plant Indus. Bul. 191, p. 33. 1910.

of each of the kinds so that the portions sent to the several localities should be as nearly alike as possible. The order of planting was the same at all the places, as follows: Every alternate row was planted to the Boone variety, which was used as the male parent in making hybrids. The seed of each of the four varieties, including the Boone itself, was planted in the rows alternating with the Boone.

The Boone plants standing in the alternate rows throughout the field were allowed to shed pollen. All others, including the interplanted Boone, were detasseled, so that the only pollen shed was from the Boone variety. Seed was saved from the detasseled rows only. This was of four kinds. (1) Stockton × Boone, (2) Strawberry × Boone, (3) Hickory King × Boone, and (4) cross-pollinated seed of the Boone. At each locality the experiment was placed at a distance from all other corn.

In 1913 it was planned to compare the behavior of the plants raised from the seed produced at the three localities when grown at each of these places. Although grown in the same field, no attempt was made to compare the relative merits of the several hybrids, each hybrid together with the pure-seed Boone constituting a separate experiment involving only the comparison of the yield from the seed of the three localities. Thus, there were four experiments to be made at each of the three places. Since the arrangement was the same at all places, one description will suffice.

To compare the Stockton × Boone hybrid from the three localities, the seed from the several places were planted in adjoining rows, the first row from the Kansas seed, the second from the Texas seed, and the third from the Maryland seed. The series was repeated 10 times, making 10 distinct comparisons. A similar procedure was followed with the three other hybrids and with the pure-seed Boone.

At Stockton, Kans., excessive drouth destroyed the entire corn crop. Since no results were secured from Stockton, the behavior of the Kansas-grown seed will be eliminated from the discussion of the results, which will be confined, therefore, to the experiments conducted in Texas and Maryland.

At Victoria, Tex., the rows were 100 feet long. The seed was drilled, and the plants were thinned to about 2 feet in the row. When harvested, a weighed sample of 20 pounds of ears was saved from each row. This sample was thoroughly air-dried, after which it was again weighed to determine the loss of water. The percentage of grain to cob was also determined. No significant differences in water content or percentage of grain were found in the crops from the seed from different localities, and these determinations are, therefore, not discussed.

At Lanham, Md., the seed was planted in hills 3 feet apart, in rows 132 feet long. The plants were thinned to one stalk per hill. The method of harvesting was similar to that of Victoria except that no determinations of dry weight were made.

At each locality the corn from all the experiments was harvested the same day; and the weight of ears, together with the number of plants, was recorded for each row.

To avoid, so far as possible, differences due to inequalities of soil and to obtain reliable averages, each pair of rows consisting of one row each of Maryland- and Texas-grown seed was considered a separate test. The relative behavior of the plants from the two sources of seed was determined by an average of all the comparisons, usually 10 in number.

In Table I are given the yield in pounds per row and the yield per plant from the several rows. Yields which stand opposite in the table are from adjoining rows in the field.

TABLE I.—*Behavior of plants from Maryland- and Texas-grown seed subsequently planted in Maryland and Texas*

Variety and factor.	Compared at Lanham, Md.			Compared at Victoria, Tex.		
	Yield from seed produced at Lanham.	Yield from seed produced at Victoria.	Yield from seed produced at Lanham expressed as a percentage of the mean.	Yield from seed produced at Lanham.	Yield from seed produced at Victoria.	Yield from seed produced at Lanham expressed as a percentage of the mean.
STOCKTON × BOONE						
	Pounds.	*Pounds.*	*Per cent.*	*Pounds.*	*Pounds.*	*Per cent.*
Yield per plant	0.90	1.15	87.8	0.65	0.65	100.0
Do	.90	1.06	91.8	.65	.66	99.2
Do	.92	.93	99.4	.62	.56	105.1
Do	.93	.81	106.9	.63	.81	87.5
Do	.89	.74	109.2	.65	.63	101.5
Do	.60	.52	107.1	.62	.69	94.6
Do	.26	.61	59.8	.64	.66	98.5
Do	.58	.56	101.7	.59	.62	97.5
Do	.78	.78	100.0	.64	.64	100.0
Do	.67	.71	97.1	.64	.66	98.5
Average	.743	.787	96.08±2.75	.633	.658	98.25±0.86
Yield per row	30.5	31.0	99.1	32.5	33.0	99.2
Do	32.5	41.5	87.8	31.8	32.5	98.9
Do	33.0	32.5	100.8	32.0	32.0	100.0
Do	25.0	29.0	92.6	32.8	38.0	92.6
Do	29.5	18.5	122.9	34.0	31.5	103.8
Do	9.0	11.0	90.0	28.5	36.5	87.7
Do	5.5	19.5	44.0	34.5	30.5	106.2
Do	18.0	18.5	98.6	31.0	36.5	91.9
Do	23.5	26.5	94.0	34.8	34.5	100.4
Do	22.0	22.0	100.0	38.0	33.0	107.0
Average	22.85	25.0	92.99±3.25	32.97	33.80	98.76±1.64
STRAWBERRY × BOONE						
Yield per plant	1.80	1.92	96.8	0.70	0.51	115.7
Do	1.81	1.61	105.9	.72	.72	100.0
Do	1.92	1.74	104.9	.78	.84	96.3
Do	1.89	1.64	107.1	.80	.84	97.6

TABLE I.—*Behavior of plants from Maryland- and Texas-grown seed subsequently planted in Maryland and Texas*—Continued

STRAWBERRY × BOONE—continued

Variety and factor.	Compared at Lanham, Md.			Compared at Victoria, Tex.		
	Yield from seed produced at Lanham.	Yield from seed produced at Victoria.	Yield from seed produced at Lanham expressed as a percentage of the mean.	Yield from seed produced at Lanham.	Yield from seed produced at Victoria.	Yield from seed produced at Lanham expressed as a percentage of the mean.
	Pounds.	*Pounds.*	*Per cent.*	*Pounds.*	*Pounds.*	*Per cent.*
Yield per plant....	1.83	1.65	105.2	0.74	0.82	94.9
Do.....	1.72	1.38	111.0	.74	.85	93.1
Do.....				.77	.74	102.0
Do.....				.69	.78	93.9
Do.....				.71	.65	104.4
Do.....				.70	.72	98.6
Average....	1.83	1.66	105.15±1.09	.735	.747	99.65±1.32
Yield per row.....	75.5	71.0	103.07	39.00	27.30	117.7
Do.....	63.5	64.5	99.22	42.50	30.30	116.8
Do.....	71.0	61.0	107.58	41.50	32.00	112.9
Do.....	68.0	62.5	104.21	39.00	34.50	106.1
Do.....	62.0	61.0	100.81	37.00	37.50	99.3
Do.....	67.0	47.0	117.54	36.80	37.50	99.0
Do.....				39.30	33.30	108.3
Do.....				37.00	39.80	96.4
Do.....				37.00	30.50	109.6
Do.....				38.00	38.00	100.0
Average....	67.8	61.2	105.40±1.80	38.70	34.05	106.61±1.82

HICKORY KING × BOONE

Yield per plant....	0.63	0.58	104.1	0.71	0.57	110.9
Do.....	.51	.60	91.9	.60	.58	101.7
Do.....	.53	.60	93.8	.57	.57	100.0
Do.....	.86	.97	94.0	.62	.57	103.9
Do.....	1.18	1.15	101.3	.58	.53	104.5
Do.....	1.25	1.08	107.3	.62	.57	104.2
Do.....	1.29	1.50	92.5	.56	.43	113.1
Do.....	1.37	1.24	105.0	.56	.57	99.1
Do.....	1.20	1.36	93.8	.63	.51	110.5
Do.....	1.34	1.30	101.5	.61	.55	105.2
Average....	1.016	1.038	98.22±1.42	.606	.545	105.31±1.0
Yield per row.....	21.5	21.0	101.2	31.00	21.00	119.2
Do.....	17.5	18.5	97.2	29.00	23.00	111.5
Do.....	17.5	18.0	98.6	28.00	25.00	105.7
Do.....	32.0	40.5	88.3	32.30	27.30	108.4
Do.....	46.0	38.0	109.5	31.30	27.50	106.4
Do.....	50.0	40.0	111.1	28.00	26.00	103.7
Do.....	51.5	58.5	93.6	28.80	23.00	111.1
Do.....	52.0	46.0	106.1	32.80	24.00	115.4
Do.....	47.0	49.0	97.9	31.50	25.50	110.5
Do.....	51.0	52.0	99.0	28.50	26.50	103.6
Average....	38.6	38.15	100.25±1.43	30.10	24.87	109.55±1.13

TABLE I.—*Behavior of plants from Maryland- and Texas-grown seed subsequently planted in Maryland and Texas*—Continued

BOONE

Variety and factor.	Compared at Lanham, Md.			Compared at Victoria, Tex.		
	Yield from seed produced at Lanham.	Yield from seed produced at Victoria.	Yield from seed produced at Lanham expressed as a percentage of the mean.	Yield from seed produced at Lanham.	Yield from seed produced at Victoria.	Yield from seed produced at Lanham expressed as a percentage of the mean.
	Pounds.	*Pounds.*	*Per cent.*	*Pounds.*	*Pounds.*	*Per cent.*
Yield per plant	1.38	1.36	100.7	0.56	0.78	83.6
Do	1.15	1.21	97.5	.60	.73	90.2
Do	1.08	1.36	88.5	.58	.68	92.1
Do	1.30	1.26	101.6	.63	.67	96.9
Do	1.10	1.13	98.7	.60	.64	96.8
Do	1.35	1.11	109.8	.69	.62	105.3
Do	1.22	1.51	89.4	.60	.68	93.8
Do	1.30	1.35	98.1	.62	.63	99.2
Do	1.16	1.22	97.5	.....	.....	.....
Do	1.14	1.24	95.8	.....	.....	.....
Average	1.218	1.275	97.76±0.91	.610	.678	94.74±1.54
Yield per row	41.5	45.0	95.95	30.50	27.00	106.1
Do	42.5	46.0	96.05	29.50	32.30	95.6
Do	45.5	47.5	97.85	33.50	32.00	102.3
Do	53.5	48.0	105.42	34.00	30.00	106.3
Do	39.5	43.0	95.76	34.50	30.00	107.0
Do	44.5	41.0	104.09	32.00	29.30	104.7
Do	47.5	53.0	94.53	32.50	28.00	107.4
Do	48.0	46.0	102.13	31.25	32.00	98.8
Do	44.0	47.5	96.17	.....	.....	.....
Do	41.0	49.5	90.61	.....	.....	.....
Average	44.75	46.65	97.87±1.02	32.22	30.06	103.5±1.50

The yield of the plants from the Maryland- and Texas-grown seed is made comparable by expressing the yield of the former as a percentage of the mean yield of both. For example, in Table I where the behavior of the Stockton × Boone hybrid is considered, the yield per plant of the first row at Lanham, Md., which was from Maryland-grown seed, is shown as 0.90 pound; the adjoining row from Texas-grown seed yielded 1.15 pounds per plant. The mean of the yield of these two rows is 102.5 pounds, and the yield from the Maryland-grown seed is 87.8 per cent of this mean, the value given in the fourth column. The average of the 10 comparisons is, in this case, 96.08 per cent—that is, the yield of the plants from the Maryland-grown seed averages 3.92 per cent below the mean yield of this strain grown in Maryland. This expression for the relative behavior in Maryland of the plants from the Maryland-grown seed is to be compared with the results of the similar comparison made in Texas, given in the next three columns. From these it is seen that in Texas the yield per plant of the plants from Maryland-grown seed was 98.25 per cent of the mean yield of the strain.

At both localties the Maryland-grown seed of this cross is inferior to that produced in Texas, but the point to which attention is now directed is that the inferiority is greater in Maryland than in Texas.

Table II contrasts the average behavior of each of the three hybrids and the pure-seed Boone at the two localities. For example, when compared in Maryland, the yield per row of plants from the Maryland-grown seed of Stockton × Boone averaged 92.99 per cent of the mean yield of the cross. In Texas the same comparison showed the average yield of plants from Maryland-grown seed to be 98.76 per cent of the mean. Thus, the plants from Maryland-grown seed averaged 5.77 per cent higher in yield in Texas than in Maryland.

TABLE II.—*Average behavior of Maryland-grown seed expressed as a percentage of the mean of Maryland- and Texas-grown seed, 1913*

Factor and kind of seed.	Compared in Maryland.	Compared in Texas.	Comparative increase of Maryland-grown seed in Texas.	Increase divided by probable error.
Yield per plant:	*Per cent.*	*Per cent.*	*Per cent.*	*Per cent.*
Stockton × Boone	96.08±2.75	98.25±0.86	2.17±2.88	0.75
Strawberry × Boone	105.15±1.09	99.65±1.32	−5.50±1.71	−3.22
Hickory King × Boone	98.22±1.42	105.31±1.05	7.09±1.76	4.03
Boone	97.76±0.91	94.74±1.54	−3.02−1.69	−1.69
Yield per row:				
Stockton × Boone	92.99±3.25	98.76±1.64	5.77±3.64	1.59
Strawberry × Boone	105.40±1.80	106.61±1.82	1.21±2.56	.47
Hickory King × Boone	100.25±1.43	109.55±1.13	10.01±1.58	6.34
Boone	97.87±1.02	103.50±1.50	5.63±1.81	3.11

In comparing the yields both the yield per row and the yield per plant were considered. In Texas the yield per row was much more dependent on the number of plants in the row than in Maryland. In Texas, as the number of plants increased, there was a pronounced tendency for the yield per row to increase and the yield per plant to decrease, while in Maryland an increase in the number of plants in the row was accompanied by only a small increase in the yield per row, and there was an almost corresponding increase in the yield per plant.

This difference in behavior at the two localties is not difficult to understand. In Maryland the failure to secure a perfect stand was largely the result of infertile spots in the field, and the same unfavorable conditions which reduced the number of plants also reduced the yield of those that survived. In Texas, the loss of plants was more the result of accidental factors, which influenced the yield of the remaining plants only by permitting them to take advantage of the increased space with a consequent slight increase in the yield per plant. The method of planting in Texas accentuated this difference, for with the plants close together in the row the additional space resulting from a missing plant was utilized more by the neighboring plants in the same row than by those in adjoining rows. In Maryland, on the other hand, where the plants were spaced equally in both directions, half the space made available by a missing hill would be appropriated by the adjoining rows.

TABLE III.—*Stand of plants secured from Maryland and Texas grown seed compared in Maryland and Texas*

Variety.	Compared in Maryland.			Compared in Texas.		
	Number of plants from Maryland-grown seed.	Number of plants from Texas-grown seed.	Stand of Maryland-grown seed expressed as percentage of mean.	Number of plants from Maryland-grown seed.	Number of plants from Texas-grown seed.	Stand of Maryland-grown seed expressed as percentage of mean.
Stockton × Boone	34	27	111.5	50	51	99.0
Do	36	39	96.0	49	49	100.0
Do	36	35	101.4	52	57	95.4
Do	27	36	85.7	52	47	105.1
Do	33	25	113.8	52	50	102.0
Do	15	21	83.3	46	53	92.9
Do	21	32	79.2	54	46	108.0
Do	31	33	96.9	53	59	94.6
Do	30	34	93.8	54	54	100.0
Do	33	31	103.1	59	50	108.3
Average	.....	.....	96.5±2.50	.....	.....	100.5±1.20
Strawberry × Boone	42	37	106.3	56	53	102.8
Do	35	40	93.3	59	42	116.8
Do	37	35	102.8	53	38	116.5
Do	36	38	97.3	49	41	108.8
Do	34	37	95.8	50	46	104.2
Do	39	34	106.8	50	44	106.4
Do	.....	.....	.....	51	45	106.3
Do	.....	.....	.....	54	51	102.9
Do	.....	.....	.....	52	47	105.1
Do	.....	.....	.....	54	53	100.9
Average	.....	.....	100.4±1.86	.....	.....	107.1±1.18
Hickory King × Boone	34	36	97.1	44	37	108.6
Do	34	31	104.6	48	40	109.1
Do	33	30	104.8	49	44	105.4
Do	37	42	93.7	52	48	104.0
Do	39	33	108.3	54	52	101.9
Do	40	37	103.9	45	46	98.9
Do	40	39	101.3	51	54	97.1
Do	38	37	101.3	58	42	116.0
Do	39	36	104.0	50	50	100.0
Do	38	40	97.4	47	48	98.9
Average	.....	.....	101.6±.98	.....	.....	104.0±1.31
Boone	30	33	95.2	54	36	120.0
Do	37	38	98.7	49	44	105.4
Do	42	35	109.1	58	47	110.5
Do	41	38	103.8	54	45	109.1
Do	36	38	97.3	57	47	109.6
Do	33	37	94.3	46	47	98.9
Do	39	35	105.4	54	41	113.7
Do	37	34	104.2	50	51	99.0
Do	38	39	98.7	.....	.....	.....
Do	36	40	94.7	.....	.....	.....
Average	.....	.....	100.1±1.24	.....	.....	108.3±1.72

Ability to produce a stand may legitimately be considered one of the manifestations of greater vigor. That it is a definite and positive factor is shown in Tables III and IV, in which the relative stands are compared. With all four kinds a comparison of the relative stand at the two localities is in favor of the transferred seed. In the Boone variety the transfer of seed has resulted in an 8 per cent increase of stand, a difference nearly four times the probable error. Since the analysis of the comparative stand of local and transferred seed shows that the differences are not accidental, but are consistently in favor of the transferred seed, it would seem that yield per row is a more reliable measure of comparative vigor than yield per plant. Yield per row is the measure of the practical results, and from this standpoint it is seen that all four strains showed an increase in yield as a result of transfer of seed. In Texas, where there was a definite tendency for an increased number of plants in a row to reduce the yield per plant, yield per plant is obviously ill calculated to bring out the real difference in vigor.

TABLE IV.—*Average stand of plants secured from Maryland-grown seed expressed as a percentage of the mean of Maryland and Texas-grown seed*

Kind of seed.	Compared in Maryland.	Compared in Texas.	Increased stand of Maryland-grown seed in Texas.	Increase divided by probable error.
	Per cent.	*Per cent.*	*Per cent.*	*Per cent.*
Stockton × Boone	96. 5±2. 50	100. 5±1. 20	4. 0±2. 77	1. 44
Strawberry × Boone	100. 4±1. 86	107. 1±1. 18	6. 7±2. 20	3. 04
Hickory King × Boone	101. 6±0. 98	104. 0±1. 31	2. 4±1. 63	1. 47
Boone	100. 1±1. 24	108. 3±1. 72	8. 2±2. 12	3. 87

EXPERIMENTS IN 1915 AND 1916

The results of the 1912 and 1913 experiments were so at variance with current belief that it was thought best to obtain additional evidence before publishing. A somewhat similar experiment was therefore planned and carried out during the years 1915 and 1916. The same varieties were used as in the previous experiment, but the localities were changed by substituting Greenville, Tex., and Sacaton, Ariz., for Victoria, Tex., and Stockton, Kans.

Crop failure at Greenville again limited the experiment to two localities: Lanham, Md., and Sacaton, Ariz. At Sacaton the temperatures are high, and there is practically no rainfall during the growing season, the crop being grown by means of irrigation.

To eliminate differences due to irregularities in the stand of plants, a different system of planting was adopted. Seed from both localities were planted in each hill, the seed from the two sources being identified by their positions in the hill. At harvest the measurements were confined to the hills which contained plants from both Maryland- and Ari-

zona-grown seed. For all such hills the height of each plant was recorded with the total length of the ear or ears. In this way each hill constituted a unit of comparison. The height of each plant was expressed as a percentage of the mean height of the two plants in the same hill. These determinations were then averaged to secure an expression of the mean behavior of the plants from each source of seed. Length of ear was treated in the same way. Table V gives the results. Unfavorable conditions so reduced the yields at Lanham, Md., that length of ear was recorded for only three of the strains, and even for these there was so much variation that the results are of doubtful significance. They serve, however, to supplement the results on the height, with which they are in accord.

TABLE V.—*Average behavior of Maryland-grown seed expressed as a percentage of the mean of Maryland- and Arizona-grown seed, 1916*

Factor.	Kind of seed.		Compared in Maryland.	Compared in Arizona.	Increase of Maryland-grown seed in Arizona.	Increase divided by probable error.
	Produced in Maryland, 1915.	Produced in Arizona, 1915.				
			Per cent.	*Per cent.*	*Per cent.*	*Per ct.*
Height of plants.	Stockton×Boone.....	Stockton×Boone....	92.9±1.21	98.9±0.38	6.0±1.27	4.72
Do....	Boone×Stockton.....	.....do.....	92.7±0.85	99.7±0.43	7.0±0.95	7.37
Do....	Strawberry×Boone..	Strawberry×Boone.	89.2±1.74	97.1±0.58	7.9±1.83	4.32
Do....	Boone×Strawberry..	.....do.....	95.7±1.44	100.9±0.46	5.2±1.50	3.47
Do....	Hickory King × Boone.	Hickory King × Boone.	103.5±0.53	97.5±0.46	−6.0±0.70	−8.57
Do....	Boone×Self.....	Boone×Boone.....	108.1±0.90	109.0±0.56	0.9±1.06	0.85
Length of ear.	Boone×Stockton.....	Stockton×Boone....	88.8±7.6	103.9±2.7	15.1±8.1	1.9
Do....	Hickory King × Boone.	Hickory King × Boone.	112.4±2.2	105.1±3.3	−7.3±4.0	1.8
Do....	Boone×Self.....	Boone×Boone.....	146.3±6.0	159.4±2.9	13.1±5.7	2.3

At Sacaton, Ariz., in 1915 reciprocal crosses were made with Stockton × Boone and Strawberry × Boone, and these reciprocals were separately compared with the seed grown at Lanham, where Boone was used only as the male parent. In all of the six comparisons except one, transferring the seed resulted in increased height; and in all but one the difference is almost certainly not the result of chance.

In 1915 the crosses in both Maryland and Arizona were made by hand instead of by detasseling alternate rows as in 1912. In gathering pollen an effort was made to obtain pollen from as many plants as possible and, so nearly as might be, in equal amounts from each plant. In like manner selection of female parents was avoided so far as possible. In spite of these precautions, it is evident that there would still be a measure of selection. Some plants produce virtually no pollen, and many plants fail to develop an ear. Furthermore, since the plants were thinned to a stand of one in a hill from each locality, obviously weak plants being removed, it would seem that here too there would be a tendency to retain

the types of plants best adapted to the conditions where the experiment was tried. The entire effect of selection would be to favor the home-grown seed, and that the transferred seed was not superior to the home-grown in every instance may not be held to vitiate the cases in which significant differences in favor of the transferred seed were observed.

The results indicate, however, that the stimulation is more pronounced in some stocks than in others. Thus, in the 1916 comparisons Boone × Hickory King stands out as a conspicuous exception. In all other stocks the transferred seed produced taller plants than the home-grown seed; but with Boone × Hickory King, the home-grown seed exceeded the transferred by 6.2 per cent, a difference not to be ascribed to chance, being more than eight times the probable error. Of the three stocks in which the yield was taken, Boone × Hickory King is also the only one to show superiority for the home-grown seed. Taken alone, the differences in yield could not be considered significant, but the agreement with the results for height confirms the reliability of these results.

The insignificant increase in the case of Boone may be explained by the fact that the Arizona-grown seed was more closely selected to fit the Arizona conditions than were the other kinds. At Sacaton the pure-seed Boone was obtained by selfing. This procedure would restrict the plants from which seed was secured to those able to produce both ears and pollen under Arizona conditions. With cross-pollinated seed and hybrids, plants that produced no ears would be represented in the progeny as male parents.

DISCUSSION OF RESULTS

Three classes or degrees of new-place effects ("neotopism") have been recognized by Cook[1], chiefly with reference to cotton: (1) Those in which there is merely a stimulation of growth; (2) those in which there is also a definite general change of the hereditary characteristics of the variety; and (3) those in which the new conditions call forth a promiscuous mutative diversity.

The results here reported give evidence under the first of these categories only. With respect to the more pronounced changes that follow the transfer of varieties from the Tropics to a temperate climate it may be said that many such changes do occur in maize, some of which at least are inherited. For conclusive qualitative evidence on this point, however, there is lacking definite information regarding the behavior of the introduced varieties in their native countries.

Roberts[2] has pointed out that the striking effects which have been ascribed to acclimatization in maize are to be referred either to cross-pollination with native varieties or to the results of selection. The effect of cross-pollination, which misled early investigators, has presumably

[1] COOK, O. F. ASPECTS OF KINETIC EVOLUTION. *In* Proc. Wash. Acad. Sci., v. 8, 1906, p. 236. 1907.

[2] ROBERTS, H. F. ACCLIMATIZATION WITH REFERENCE TO CORN BREEDING. *In* 1st Ann. Rpt. Kans. Corn Breeders' Assoc., [1905]/06, p. 60–64. 1906.

been eliminated from recent experiments, but the effects of selection are so pronounced and speedy that in experiments hitherto reported any direct effect of the environment on the characters of the plants would be completely masked. The characteristics of a maize variety are altered readily by selection. When grown in a new locality for a few years, even without conscious selection, the type may change rapidly; and when brought back to the original locality, it is in reality a different variety. The characters brought into prominence in the new locality may render the stock less suited to the old conditions, though better adapted to the new.

It would be very difficult, if not impossible, to eliminate completely all selective action. Even when all seed is saved, those individuals or types of plants which are best adapted to the conditions under which they are grown will produce a greater proportion of the seed than will the types which are less well adapted, and those least adapted may produce no seed at all. In two localities where different conditions prevail, the highest yielding plants—hence those contributing the largest proportion of the seed—would presumably be of different types; and when brought together and compared, we should expect to find a slight advantage for the locally grown seed. Yet the results of the present experiments indicate that the effect of selection during a single season may be so slight as not to mask completely the opposing new-place effect.

Since new-place effect in maize seems to operate as a stimulus, it would tend to obscure any lack of adaptation in newly introduced varieties. The recognition of new-place effect may be said, therefore, to increase rather than diminish the importance that must be assigned to adaptation.

As a result of the stimulation due to new-place effect, the cultivation of an inferior strain might be extended as a result of its satisfactory performance the first year following its introduction.

The stimulus that followed the transfer of seed in these experiments is doubtless similar to the increased vigor imparted to many vegetables by growing the crop in localities remote from the place where the seed was produced. The economic utilization of increased vigor secured in this way is usually confined to crops which are grown for the sake of some part other than the seed. In cotton, for example, the increase of vigor in the plants following a transfer of seed is often very pronounced, although the crop of seed and fiber may be reduced. In maize, as a result of the determinate habit of the plant, vegetative vigor and seed production are more closely associated, so that the possibility of practical utilization seems greater.

CONCLUSIONS

Hybrids between the same pairs of varieties made at different localities showed no decrease in yield as a result of transferring the first-generation seed to a new locality. On the contrary, the change of

environment seemed to act as a stimulus, with the result that the yields were increased in all but one of the hybrids tested. One unhybridized variety was included in the experiment, and this also gave slightly increased yields as a result of being transferred to a new environment.

In 6 of the 10 comparisons the increase is too large to be ascribed to experimental error and indicates that new-place effect should be taken into consideration as a factor of production.

That significant increases may be secured by taking advantage of new-place effect in maize should not be used as an argument in favor of the general transfer of seed. There is no evidence that the importance of using acclimatized seed has been overestimated. On the contrary, the experiments show that new-place effect may often obscure the differences between acclimatized and unacclimatized seed when first compared, and thus interfere with a full appreciation of the value of adaptation.

The investigations show the existence of a hitherto-neglected factor in maize production, but much more extensive experiments are needed to ascertain the extent and practical importance of this factor. The existence of one definite exception indicates that the tendency to increased vigor following a transfer of seed is not universal. The results also indicate that adaptation in maize comes about through selection rather than as a direct reaction to the environmental conditions.

RELATION OF THE VARIABILITY OF YIELDS OF FRUIT TREES TO THE ACCURACY OF FIELD TRIALS[1]

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INTRODUCTION

The value of the outcome of any trial depends upon the probability that a similar result will be obtained if the trial is repeated. In recent years the agricultural experiment stations of all countries have greatly increased the number and size of their field trials. A casual examination of such trials usually shows a wide range in the reliability of the results. The yields of control plots in different parts of the same tract will often differ as much among themselves as the yields of fertilized and unfertilized plots differ from each other. The purpose of this paper is to present the results of a study of the variation in recorded yields of fruit trees taken singly and in groups of various sizes, and especially to determine the effect upon variability of various combinations and repetitions of unit plots.

It is recognized that the results of a single experiment are often untrustworthy; yet experimenters have published single results and have based practical advice upon them. However well planned field trials may be, the interpretations of the results can hardly be considered of infinite reliability—that is, results which will invariably be obtained when the trials are repeated. The best that can be done is to construct the most probable results from the more or less varying observed results of individual trials. If it is impossible to obtain perfect accuracy, it is not impossible to fix the limits of error and thus to determine whether the differences obtained are due to the treatments applied or to unavoidable errors—that is, whether the differences are significant. A number of trials are necessary before a reasonably reliable result can be obtained. The average result of a series does not always represent the truth. When averages are used, they should always be accompanied by their probable errors, which are a measure of their reliability.

Before attempting to interpret the results of a plot experiment with fertilizers, it is necessary to know whether the differences observed are any greater than those which might have occurred had none of the plots been fertilized. The scientific method is to recognize the inevitable error and, while reducing it by every possible precaution, at the same time to

[1] Paper 44, University of California, Citrus Experiment Station, Riverside, Cal.

[2] The writers wish to make acknowledgment of their indebtedness for aid and criticism to Prof. H. H. Love, of Cornell University; Prof. E. B. Babcock, Dr. H. B. Frost, and other members of the University of California.

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measure its probable amount so as to make sure it is not likely to vitiate our conclusions.

One of the chief difficulties in obtaining reliable results in field trials is the natural variability of the material with which we are dealing. Crops are living organisms with inherent tendencies to vary, even though it were possible to make their environmental conditions identical.

In agronomic experiments the number of plants taken is usually so large that inherent variability ceases to be a factor of importance. In horticultural experiments, however, where fruit trees are under observation, the limited number of trees possible to include in a plot may make the factor of inherent variability an important consideration.

Further variation is induced as a result of the many factors of the environment which are beyond the control and possibly the recognition of the experimenter. Some of these factors are independent; others react upon one another. In designing a set of field trials, we try to avoid, as far as possible, all secondary factors which may exert a disturbing influence.

Lack of uniformity in both the physical and chemical characteristics of the soil is one of the foremost factors causing variation in productivity of plants. Apparently uniform surface soils may be underlain with a heterogeneous subsoil. Differences also occur which are not evident on a careful inspection of both the soil and the crops, but which are easily measured by weighing the yields. In other words, the weighing machine is more sensitive than the eye and reveals differences that mere inspection can not detect.

The past treatment of the soil brings in variables the significance of which may not be comprehended at the time a field trial is begun. The persistent effects left by the application of stable manure on some of the Rothamsted plots show how large a part is played by the past history of the field. Plots which for 40 years have had identical treatment still give different crop yields because of the effect of dressings of barnyard manure applied at an earlier period.

Unequal prevalence of diseases and insects may bring about further error in the results.

Besides the above sources of variation and possibly outweighing them at times is the effect of season. No season is entirely normal; and it is only when the experiment has been repeated for several years, or, in other words, until "a fair sample of seasons" has been made, that any sort of allowance can be made for seasonal effects.

As is shown by observations at Rothamsted, it is not possible to establish a schedule of relative yields for a series of plots, even after several years' comparison. In this case two grass plots were treated alike for 50 years; by taking the yield of one plot as the standard, the yield on the other in the same season has been as low as 90 per cent and as high as 196 per cent.

Recent investigations, which are reviewed in succeeding pages, have thrown light upon the nature of the variability inherent in experiments conducted with groups of plants or of animals. So far as the writers know, however, few studies have yet been published upon the uncertainty with which one deals in attempting to carry on plot trials with orchard trees. In connection with plans for extending the trial plots of this Experiment Station, the writers have attempted to study the question of variability of tree yields and to formulate some plan to determine the probable reliability of the results. It is highly desirable to make the tree plots as small as possible without sacrificing too much accuracy on account of the large amount of land required for each individual.

A fruit tree is possibly more affected by environmental conditions than an annual crop growing from seed to maturity in one season. The tree roots penetrate more deeply into the soil and may be affected by soil differences to a considerable depth. Climatic conditions during the resting period may have marked influence on the crop production of the trees.

Pruning also introduces variation. Uniform pruning is desirable; yet different trees need different types of pruning, and more uniform results will be obtained if this is recognized.

Individual trees have apparent idiosyncrasies in fruiting habits. A tree may yield large crops of fruit in alternate years, with very light crops in intervening years. The character of the stock upon which the particular variety was budded to form the tree may profoundly influence the type, habit, and productivity of the adult tree. Furthermore, we can not overlook the possibility of errors in yields due to predacious animals which devour or otherwise destroy more or less fruit.

Fruit trees therefore present opportunity for more variability than would be expected in the growth of annual plants.

PREVIOUS STUDIES

The varying productivity of fruit trees has been called to our attention by the writings of Macoun (*1904*),[1] Munson (*1907*), Shamel (*1912*), Hedrick (*1912*), Stewart (*1913, p. 552–554*), Fletcher (*1913*), Coit (*1910*), Whitten (*1915*), Lewis and Vickers (*1915, p. 30–31*), Barre (*1915*), Gourley (*1915, p. 72–73*), Kraus (*1916*), and others. The above observations have been largely made in connection with plant breeding and orchard management problems, and were not made to bear necessarily on the accuracy of plot trials.

Holtsmark and Larsen (*1906*) were among the first to call attention to the errors of field trials. They recognized the inevitable variation of field results, and showed how it may be estimated by the use of the standard deviation and the coefficient of variability. They also showed

[1] Bibliographic citations in parentheses refer to "Literature cited", pp. 282–283.

that the coefficient of variability decreases as the plots are enlarged, but not proportionally to the size of the plot.

The limitations of field experiments were discussed by Carleton (*1909*), who called the attention of experimenters to the various uses of control plots, and to the general precautions necessary to obtain reliable results.

Hall (*1909*), Mercer and Hall (*1911*), and Hall and Russell (*1911*) recorded extensive studies of the soil variations in experimental grounds and the influence of size and repetition of plots upon accuracy. This work was done largely with the yields of wheat, mangel, and hay crops. The conclusions from the above work are that the error in field trials diminishes as the size of the plot increases, but that the reduction is small when the plot is enlarged to a size greater than one-fortieth of an acre. The error may be further diminished by increasing the number of plots similarly treated and scattering them about the area under experiment; but there is not much to be gained by increasing the number of plots above five.

Wood and Stratton (*1910*) sounded notes of caution concerning the interpretation of experimental results. Frequency distribution is discussed from the point of view of its bearing on the reliability of averaging results. The applications of the probable-error methods to questions of sampling for analysis, to field experiments, and to feeding experiments are illustrated. The probable error of field experiments was investigated by two independent methods and found to be about 5 per cent of the mean yield. Tables are given showing the number of duplicate plots or number of animals in a feeding trial which must be employed to give any desired precision in the result. It is shown that more accurate results may be obtained by employing large numbers of small scattered plots than by using one large plot.

The estimation of errors in field-plot tests has been given considerable attention by Lyon (*1912*) and coworkers. It was shown that it is not possible to establish a schedule of relative yields for a series of plots, even after several years' comparison. Also, there seems to be little gain by using plots larger than one-fiftieth of an acre in size when the comparative yield of the crops is made the criterion. An area of one-twenty-fifth of an acre of land distributed in four widely separated plots, devoted to any one test, secures a much greater degree of accuracy than the same area of land in one body. The probable error was reduced from 4.5 to 2 per cent by such distribution.

Pickering (*1911*), from studies on apples and pears, concluded that experimental plots should include 6 to 12 fruit trees. Precautionary advice was also given concerning the measurement of results by crop production, foliage, and tree characteristics. In comparing the results on the treated plots with the controls, instead of taking the average of the controls, he prefers to plot these results out and to draw a smoothed

curve through them, and then to compare the results of the experimental plots with readings taken at corresponding points of this curve.

Wood (*1911*) showed how the degree of reliance can be determined for any set of experimental results by the use of the probable error. The use of this constant was demonstrated in interpreting laboratory analyses, as well as both plot and feeding experiments. Working with mangel yields, the author calculates the number and size of plots required to attain any desired precision, and working with the probabl eerror of liveweight increase of sheep, tables are given showing the number of animals required in an experiment to attain various degrees of reliability.

Several papers by Harris (*1912*, *1913a*, *1913b*, and *1915*) have drawn our attention to several phases of the experimental error in field tests. A measure of the variability of the soil productivity is obtained by determining the correlation between the yields of ultimate small plots and the yields of various groups of adjacent plots. The more nearly this correlation approaches zero, the more homogeneous the soil. This method of measurement does not seem to provide as definite a means of obtaining a corrective term as the use of the coefficient of variability and the probable error as used by Wood, Wood and Stratton, Mercer and Hall, etc., or the contingency method of correction as used by Surface and Pearl (*1916*).

Montgomery (*1912*) has also discussed the comparative variability resulting from increasing the size of the plot and from distributing small ultimate plots over the area. The latter method was found to be more accurate. In a subsequent paper (*1913*) the relative reliability of yields of wheat planted in rows and in square blocks is discussed.

An exhaustive and discriminating discussion of the nature and magnitude of variability in the results of feeding experiments has been given by Mitchell and Grindley (*1913*). Much of their discussion is equally applicable to experimentation with plants.

Olmstead (*1914*) applied the method of least squares in calculating the reliability of the yields of the mangel and wheat crop records of Mercer and Hall, the potato records of Lyon, and the wheat yields of Montgomery. The conclusions from this series of observations are:

> The estimation of the probable error of a large number of small duplicate plots well distributed in the area devoted to a field experiment indicates that the precision of agricultural experiments can be increased by replicating the experiments on small plats.

Coombs and Grantham (*1916*) have studied the variation in the yields of rice and coconuts for one year, and discussed the range and interpretation of the probable error. They showed that the yields from any two single plots could only be significant when the difference amounted to 22.8 per cent of the mean. They also introduced calculations to show the odds that any increase is a real increase and not a probable error.

The use of controls and repeated plantings in varietal tests was studied by Pritchard (*1916*) in breeding work with sugar beets. His studies lead to the conclusion that the practice of dispensing with control rows and using the mean of all progeny rows as a standard of comparison appears to be less accurate than the employment of frequent controls. However, the employment of every alternate row as a control was not sufficient to offset the variability in yield arising from irregularities of soil.

Stockberger (*1916*) discussed the value of a number of the common methods for determining the normal yield of treated plots based upon the yields of hops. Normal yields for various plots varied widely according to the method of computation, the values in some cases differing from the actual yield by as much as 40 per cent. Repetition brought about a very marked reduction in variability, although with only five repetitions the error is still relatively large.

The work of Surface and Pearl (*1916*) shows an advance in the refinement of methods of conducting field trials. With the realization that the use of frequent control plots often produces results far from satisfactory, these workers have calculated by the contingency method the probable yield of each plot of ground in their grain-testing series. This calculated yield represents the most probable yield of each plot on the supposition that they have all been planted with a hypothetical variety whose mean yield is the same as the observed mean of the field. This "calculated" yield may then be used as a basis for determining a correction factor, whereby each area must be given a handicap plus or minus the actual yield, depending upon whether the plot in question is calculated to be a low- or a high-producing area. This method of correcting the soil variation is combined with four systematically repeated plots of one-fortieth acre of each variety, and gives a high degree of accuracy.

MATERIAL USED FOR STUDY

The studies to be reported in this paper deal with the variability of fruit-tree yields. They are based upon the individual tree yields of oranges (*Citrus sinensis*), lemons (*Citrus limonia*), walnuts (*Juglans regia*), and apples (*Malus sylvestris*) from orchards which had received uniform treatments for a number of years—indeed, so far as known, from the time of planting the trees. The orchards were carefully examined, and the records for all trees which were known to be abnormal from disease or other apparent causes were eliminated. In place of the records of trees thus eliminated the average yield of the eight surrounding trees was substituted (assuming that the tree stood at the center of a square block of nine trees).

This substitution is not entirely satisfactory, yet it was felt that it was necessary in order to compute plots of homologous size and systematic arrangement. The writers found, as a matter of fact, that there is a very high degree of correlation in these orchards between the yields

of individual trees and the average of surrounding trees. For example, they found coefficients of correlation as high as 0.652 ± 0.065 for the Eureka lemons and 0.628 ± 0.060 for the Arlington navels. According to the formula given by Harris (*1915*), the correlation between individual trees and eight adjacent trees in a plot of the Arlington navels is 0.576 ± 0.04. In view of these results, the writers felt justified in using this method of substituted values.

The fruit plantations herein discussed, to judge by the surface soil, size, and condition of the trees, as well as their apparent fruitfulness, appeal to the observer as uncommonly uniform. All the orchards studied are situated in semiarid regions and are artificially irrigated during the summer months. This fact is believed to be a distinct advantage for the purpose of reducing the variability of one year's yield compared with another, since it insures a fairly uniform water supply for the soil and reduces one of the variants inevitable in nonirrigated localities.

All yields of the several fruit and nut plantations are given in pounds per tree of the ungraded product.

DESCRIPTION OF THE PLANTATIONS

NAVEL ORANGE (ARLINGTON).—These records were of the 1915–16 yields of one thousand 24-year-old navel-orange trees near Arlington station, Riverside, Cal. The individual tree production is shown by figure 1.

The grove consists of 20 rows of trees from north to south, with 50 trees in a row, planted 22 by 22 feet. A study of the records shows certain distinct high- and low-yielding areas. The northeast corner and the south end contain notably high-yielding trees. The north two-thirds of the west side contains a large number of low-yielding trees. These areas are apparently correlated with soil variation. Variations from tree to tree also occur, the cause of which is not evident. These variations, which are present in every orchard, bring uncertainty into the results of field experiments.

In making their calculations this grove was divided by the writers into imaginary plots of any size and shape desired. The yields of these plots were then compared with one another and their variability ascertained. The distribution of both the theoretical and actual yields of this grove is shown in figure 10. The yields of the individual trees when plotted according to their frequency give a skew curve of Pearson's Type I, since the critical function

$$F = \frac{\beta_1 (\beta_2 + 3)^2}{4 (4\beta_2 - 3\beta_1) (2\beta_2 - 3\beta_1 - 6)} = -0.85.$$

The distribution of the actual yields is shown on the figure by small circles. The points for the theoretical curve were calculated by the formula

$$y = y_0 \left(1 + \frac{x}{l_1}\right)^{m^1} \left(1 - \frac{x}{l_2}\right)^{m^2}.$$

FIG. 1.—Diagram showing the individual tree yield (in pounds) of the navel-orange grove (Arlington).

NORTH.

Yield per tree (pounds).

Row.	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	Total yield per row.
1	82	77	39	114	85	60	95	94	82	71	102	108	70	108	127	64	166	156	170	83	1,953
2	74	105	78	124	81	110	131	141	122	104	117	108	116	150	131	225	199	119	180	171	2,586
3	66	69	74	79	211	89	137	23	15	107	127	163	125	140	180	168	161	203	302	277	2,716
4	75	55	84	98	132	108	147	158	101	180	83	179	105	193	148	158	106	166	255	61	2,592
5	84	71	82	109	99	127	156	110	175	159	173	196	159	39	99	152	192	268	218	142	2,810
6	48	81	51	87	69	90	136	164	112	96	46	104	136	213	197	195	191	197	184	80	2,477
7	70	81	79	109	150	90	112	121	118	158	151	116	166	131	148	180	164	182	186	156	2,668
8	54	105	82	128	101	81	135	143	198	157	211	127	148	141	115	169	180	190	238	112	2,815
9	72	82	57	86	111	75	56	153	140	107	143	180	145	161	79	75	147	156	223	172	2,420
10	28	49	48	71	84	89	33	60	165	138	126	145	149	139	107	127	152	172	32	75	1,989
11	61	31	50	65	95	75	112	112	101	120	120	131	137	217	101	123	180	124	117	171	2,243
12	68	60	100	79	98	115	110	135	74	142	90	109	175	192	182	123	173	155	35	127	2,342
13	33	80	67	69	118	64	103	190	109	210	51	78	163	116	130	101	126	121	154	69	2,152
14	36	91	107	90	80	95	78	148	107	162	161	76	123	132	114	122	152	121	167	152	2,314
15	76	109	93	89	107	49	84	164	97	124	135	147	181	183	175	90	205	167	50	146	2,471
16	49	99	81	131	109	74	123	146	156	203	146	157	156	137	180	178	140	179	159	92	2,695
17	92	123	75	45	77	61	135	93	95	224	122	178	166	170	240	208	141	168	162	90	2,665
18	34	98	89	73	123	69	139	187	164	150	163	84	111	152	176	150	150	107	130	129	2,478
19	45	57	68	37	71	83	109	45	94	139	128	141	68	79	137	90	80	130	143	107	1,851
20	53	90	128	99	72	73	80	117	137	160	152	131	116	113	169	90	57	97	110	102	2,146
21	91	80	101	117	79	135	135	62	186	132	99	80	135	134	215	105	63	85	126	69	2,229
22	73	117	83	97	93	126	116	111	115	135	105	86	103	166	164	119	115	103	152	123	2,302
23	76	83	81	84	119	67	126	96	116	111	93	44	118	165	162	34	107	81	78	28	1,809
24	148	131	86	87	98	89	107	123	108	89	57	122	102	182	135	75	65	147	86	102	2,139
25	59	66	85	71	90	58	114	90	45	116	63	99	70	117	163	22	80	71	59	59	1,597
26	86	64	78	134	174	135	111	90	93	135	112	90	152	123	149	100	33	130	114	63	2,166
27	28	76	90	144	135	111	135	116	57	190	122	128	79	165	153	164	167	195	113	89	2,457
28	51	73	79	164	161	115	127	101	71	189	180	208	140	107	154	133	107	130	103	84	2,479
29	54	82	135	141	135	90	135	160	46	120	138	90	127	82	135	63	68	91	25	5	1,922
30	162	135	155	114	163	180	209	95	163	146	164	80	110	74	54	92	90	57	103	70	2,416
31	118	147	135	120	50	95	151	61	98	148	135	189	118	130	76	82	174	88	91	107	2,313
32	103	106	62	102	135	123	135	185	148	161	180	241	151	123	95	122	135	165	137	121	2,730
33	111	69	106	158	90	104	180	145	135	117	164	180	135	161	180	135	142	160	180	90	2,742
34	135	124	135	142	140	140	239	225	193	142	156	180	218	165	213	213	180	157	152	97	3,346
35	115	100	119	164	135	169	180	196	135	112	90	159	167	90	192	139	135	135	154	69	2,755
36	127	135	116	143	135	150	214	196	169	100	180	180	164	165	225	157	195	150	251	165	3,317
37	180	104	135	148	161	135	142	164	90	154	191	135	149	116	183	173	159	210	238	107	3,074
38	118	135	135	154	141	180	160	211	161	147	198	214	200	144	215	159	77	147	214	180	3,290

39	123	159	154	181	180	159	243	293	151	148	185	194	186	200	202	112	180	156	225	164	3,585
40	184	123	239	169	150	146	211	180	178	223	213	147	162	177	145	237	192	148	204	55	3,481
41	100	168	116	225	150	179	196	214	193	218	214	167	113	117	131	101	131	127	110	85	3,074
42	202	166	132	241	210	202	201	190	144	136	155	203	168	196	121	102	167	181	120	225	3,516
43	190	168	194	239	185	176	215	249	153	163	186	175	225	190	108	191	130	155	255	186	3,747
44	208	163	225	192	225	159	180	254	149	152	140	147	155	85	122	163	154	187	166	139	3,272
45	205	201	168	188	211	249	270	238	160	198	214	238	158	87	172	244	154	258	209	168	3,913
46	136	137	182	153	229	221	261	219	129	151	137	144	201	178	152	138	218	148	171	149	3,499
47	136	205	190	177	252	197	238	341	173	138	237	211	200	246	186	246	207	254	225	167	4,270
48	195	181	260	208	212	141	288	85	123	149	237	211	216	230	151	145	91	225	225	102	3,867
49	158	227	139	114	251	206	267	282	214	135	175	230	316	299	140	146	154	135	208	117	4,089
50	142	192	140	208	210	183	243	264	161	176	226	282	275	222	180	268	194	194	283	166	4,207
Total yield per row		4,914	5,530	5,528	6,361	6,781	6,139	7,740	7,738	6,420	7,511	7,405	7,610	7,533	7,475	7,611	7,104	7,353	7,587	7,986	5,859	137,985

NAVEL ORANGE (ANTELOPE HEIGHTS).—The navel-orange grove later referred to as the Antelope Heights navels is a plantation of 480 ten-year-old trees planted 22 by 22 feet, located at Naranjo, Cal. The individual tree records of this planting are shown in figure 2 and are the yields obtained in 1916. The general appearance of the trees gives a visual impression of uniformity greater than a comparison of the individual tree production substantiates; however, the distribution of the yields approximates closely the normal curve of errors, having a skewness of only 0.001 ± 0.037, as shown by figure 11.

FIG. 2.—Diagram showing the individual tree yield (in pounds) of the navel-orange grove (Antelope Heights).

NORTH

Yield per tree (pounds)

Row.	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	Total yield per row.
1	130	200	250	250	130	300	260	285	225	255	240	225	250	200	150	3,350
2	120	150	220	230	270	275	300	340	215	195	150	250	260	250	230	3,455
3	200	150	230	300	235	280	200	170	250	250	150	150	200	250	270	3,285
4	155	205	225	295	265	320	250	130	200	200	150	210	220	180	280	3,285
5	250	225	260	300	200	290	250	185	200	155	220	165	250	255	270	3,475
6	300	200	200	240	200	210	250	220	255	150	220	135	175	255	230	3,240
7	225	250	250	225	200	275	250	200	175	150	250	170	170	260	200	3,250
8	150	325	250	225	210	225	200	250	225	150	150	205	190	265	350	3,370
9	300	150	250	290	200	100	225	225	125	225	200	250	225	200	225	3,190
10	250	185	150	175	135	170	200	225	150	200	200	165	175	225	240	2,845
11	200	100	155	170	175	200	200	210	110	300	250	190	135	205	225	2,825
12	150	200	100	230	150	200	250	215	140	200	225	160	115	220	220	2,775
13	110	150	175	135	160	180	250	220	150	200	250	225	230	225	175	2,835
14	190	200	125	165	135	150	200	190	170	200	220	300	200	225	200	2,870
15	200	150	140	130	100	185	220	150	150	150	275	200	190	205	200	2,645
16	210	150	120	100	110	150	150	225	160	150	175	125	150	150	220	2,345
17	140	100	120	115	135	160	250	200	140	150	150	225	145	125	165	2,320
18	105	100	100	200	150	200	200	185	110	125	125	150	100	200	150	2,200
19	120	150	50	150	100	200	200	225	140	200	125	125	200	150	150	2,285
20	130	120	75	120	150	200	150	225	100	150	75	200	200	175	75	2,145
21	150	100	125	200	175	275	160	200	100	100	100	200	150	75	100	2,210
22	195	105	150	150	185	200	190	160	55	150	100	225	100	150	100	2,215
23	100	110	130	155	265	150	200	140	90	150	100	150	110	100	125	2,075
24	100	150	80	200	150	175	175	125	75	100	200	240	90	135	130	2,125
25	105	100	150	150	175	200	175	150	150	175	200	150	150	150	100	2,280
26	100	100	150	270	100	150	175	250	175	175	250	150	125	150	100	2,420
27	100	150	175	230	150	150	150	270	190	200	225	200	150	200	190	2,730
28	135	185	180	280	250	115	180	175	75	200	175	185	150	150	200	2,635
29	245	255	100	250	300	260	185	150	150	250	175	200	210	125	150	3,005
30	200	170	150	50	350	240	240	200	220	210	215	250	225	200	250	3,170
31	225	225	180	250	115	235	225	200	250	270	235	200	185	150	165	3,110
32	200	200	185	200	215	240	200	200	190	250	250	250	175	120	150	3,025
33	250	150	185	150	170	155	220	250	220	250	250	250	100	250	275	3,125
Total yield per row	5,740	5,460	5,385	6,580	6,010	6,815	6,930	6,745	5,330	6,235	6,275	6,475	5,700	6,175	6,260	92,115

VALENCIA ORANGE.—The Valencia orange grove is composed of 240 15-year-old trees, planted 21 feet 6 inches by 22 feet 6 inches, located at Villa Park, Cal. Figure 3 represents this planting and the individual tree yields which were obtained in 1916.

EUREKA LEMON.—The lemon yields were obtained from a grove of 364 23-year-old trees, located at Upland, Cal. Figure 4 represents the individual tree yields of this planting. The records extend from October 1, 1915, to October 1, 1916. The grove consists of 14 rows of 23-year-old trees, extending north and south, with 26 trees in a row, planted 24 by

FIG. 3.—Diagram showing the individual tree yield (in pounds) of the Valencia orange grove.

NORTHWEST

Yield per tree (pounds)

Row.	1	2	3	4	5	6	7	8	9	10	11	12	Total yield per row
1	100	375	250	275	275	150	200	175	225	275	200	125	2,625
2	375	325	275	375	200	375	225	230	275	225	225	262	3,367
3	37	75	175	260	150	300	25	275	450	250	325	250	2,572
4	63	175	250	350	300	275	325	300	250	275	275	275	3,113
5	75	75	175	300	300	325	300	275	375	275	200	275	2,950
6	100	212	125	25	200	275	325	250	250	100	150	75	2,087
7	50	136	100	75	250	100	250	100	190	150	225	225	1,851
8	300	200	100	75	150	200	200	275	200	200	150	250	2,200
9	100	175	50	300	263	363	350	300	337	338	375	150	3,101
10	100	150	100	150	150	262	250	275	363	337	350	250	2,737
11	75	125	125	275	125	275	150	225	312	188	225	150	2,250
12	225	225	350	250	300	150	300	300	262	265	225	225	3,077
13	425	200	225	325	75	275	425	275	375	300	25	225	3,150
14	50	350	200	125	225	150	225	250	250	300	300	175	2,600
15	437	238	225	200	350	375	225	225	300	400	375	350	3,700
16	400	163	150	300	450	300	400	350	350	250	325	225	3,663
17	350	275	400	300	462	288	412	363	375	300	350	100	3,975
18	175	325	250	350	350	350	100	300	300	200	175	175	3,140
19	350	350	400	250	275	275	212	188	350	250	150	100	3,150
20	400	350	450	375	200	300	350	375	275	350	325	100	3,850
Total yield per row	4,087	4,499	4,375	4,935	5,050	5,363	5,249	5,306	6,064	5,318	4,950	3,962	59,158

FIG. 4.—Diagram showing the individual tree yield (in pounds) of the Eureka lemon grove.

NORTH

Yield per tree (pounds)

Row.	1	2	3	4	5	6	7	8	9	10	11	12	13	14	Total yield per row.
1	259	258	188	251	379	325	201	232	349	237	296	264	241	104	3,584
2	204	157	227	314	381	264	283	211	292	329	273	218	236	222	3,611
3	475	235	305	183	412	251	210	271	443	348	378	298	310	222	4,341
4	383	292	274	263	288	356	260	172	327	322	345	295	279	375	4,231
5	510	250	190	202	313	300	157	169	327	399	281	304	321	280	4,003
6	421	188	277	338	369	346	286	140	382	346	233	245	313	333	4,217
7	425	228	167	195	343	363	202	167	334	377	312	300	308	266	3,987
8	218	274	206	133	234	341	211	169	386	429	298	282	277	100	3,557
9	301	257	176	276	230	312	167	206	180	471	342	238	339	342	3,837
10	409	264	164	188	221	319	234	149	439	354	248	279	293	287	3,848
11	152	165	123	194	276	336	278	177	314	441	283	281	282	279	3,581
12	79	118	150	120	258	222	261	245	268	308	278	322	306	222	3,157
13	287	196	170	264	292	241	235	159	341	399	317	307	298	295	3,801
14	348	258	161	267	345	345	237	216	380	353	365	301	200	285	4,061
15	81	199	144	180	268	278	213	209	336	312	299	279	223	248	3,269
16	250	146	106	282	253	290	245	218	360	368	322	338	344	245	3,767
17	111	165	223	259	227	361	278	116	304	315	338	318	326	311	3,652
18	195	162	135	241	275	291	256	122	259	363	385	358	356	294	3,692
19	333	156	170	154	357	325	186	80	226	335	369	358	223	288	3,560
20	291	172	127	203	104	239	222	112	364	371	349	402	276	338	3,570
21	111	189	181	255	332	328	235	176	298	398	329	307	204	244	3,587
22	361	114	127	296	326	252	396	158	282	325	294	322	287	236	3,776
23	210	245	184	315	414	294	201	128	416	313	326	410	223	276	3,954
24	140	197	245	307	350	388	343	289	410	302	341	283	253	292	4,040
25	126	229	216	185	275	297	412	241	328	262	333	280	131	277	3,592
26	241	263	189	220	397	399	341	335	438	266	413	237	328	316	4,383
Total yield per row	6,921	5,377	4,825	5,985	7,919	8,063	6,550	4,867	8,783	9,043	8,347	7,825	7,176	6,977	98,658

24 feet apart. This grove presents the most uniform appearance of any under consideration. The land is practically level, and the soil is apparently uniform in texture. The records show a grouping of several low-yielding trees; yet a field observation gives one the impression that the grove as a whole is remarkably uniform.

SEEDLING WALNUTS.—The walnut-tree records used in the following calculations were obtained during the seasons of 1915 and 1916 from a 24-year-old Santa Barbara softshell seedling grove, located at Whittier, Cal. The planting is laid out 10 trees wide and 32 trees long, entirely surrounded by additional walnut plantings, except on a part of one

FIG. 5.—Diagram showing the individual tree yield (in pounds) of the seedling walnut orchard.

NORTH

Yield per tree (pounds)

Row.	1	2	3	4	5	6	7	8	9	10	Total yield per row.
5	8	88	185	113	113	167	87	188	164	164	1,277
6	172	119	44	111	35	33	67	207	164	164	1,116
7	164	99	135	40	207	94	79	136	195	97	1,246
8	74	102	96	80	78	57	104	104	54	62	811
9	122	22	80	108	112	118	80	144	52	58	896
10	120	71	103	74	129	40	123	40	107	21	828
11	160	111	94	135	121	131	84	126	30	24	1,016
12	170	135	128	82	93	93	93	72	8	96	970
13	95	89	71	66	61	134	61	70	60	64	771
14	65	50	61	195	89	45	61	43	25	43	677
15	85	101	100	100	72	63	133	14	73	101	842
16	78	106	88	112	76	86	15	113	53	83	815
17	51	34	101	71	82	99	15	81	28	50	612
18	17	104	25	95	57	76	96	69	25	65	629
19	75	69	73	68	6	35	90	80	94	13	603
20	117	179	123	35	47	51	104	85	42	75	858
21	91	188	88	105	65	86	91	31	72	97	914
22	122	122	103	180	120	165	144	106	85	105	1,252
23	99	144	69	124	48	197	122	77	117	83	1,080
24	83	155	170	32	142	97	101	160	90	87	1,117
25	44	83	198	192	113	76	81	131	65	39	1,022
26	86	176	125	130	147	119	210	113	149	131	1,386
27	71	50	151	58	112	71	104	94	113	126	950
28	87	111	140	170	123	96	160	109	95	148	1,169
29	41	136	74	54	115	99	130	87	166	129	1,031
30	86	158	95	80	75	178	90	115	150	128	1,155
31	169	150	151	138	148	151	73	94	166	235	1,475
32	112	100	166	188	39	192	157	215	160	201	1,538
Total yield per row	2,664	3,052	3,037	2,936	2,625	2,849	2,755	2,904	2,545	2,689	28,056

side which is adjacent to an orange grove. The trees are planted on the square system, 50 feet apart. Figure 5 gives the yield and arrangement of these trees.

JONATHAN APPLES.—The apple records [1] were obtained from a 10-year old Jonathan apple orchard located at Providence, Utah. The surface soil of this orchard is very uniform to all appearances except on the extreme eastern edge, where the percentage of gravel increases slightly. The trees are planted 16 feet apart, east and west, and 30 feet apart north and south. Figure 6 gives the yield and arrangement of these trees.

[1] The authors wish hereby to express their appreciation of the kindness of the Utah Experiment Station in furnishing these records.

RANGE OF INDIVIDUAL TREE YIELDS

The extremes of individual tree productivity are shown in Table I. This indicates a wide range of variation even in oranges, lemons, and apples, which are clonal varieties. The greatest range, however, is in the yield of the seedling walnuts. The variations set forth in this table may seem excessive to workers with annual crops. To those familiar with the variation in tree crops, however, this will be recognized as only the normal variation which occurs in most fruit plantations growing on apparently uniform soil, as mentioned in a previous section. Wide variation in the vigor of the rootstock, as well as variation in soil pro-

FIG. 6.—Diagram showing the individual tree yield (in pounds) of the Johnathan apple orchard.

Yield per tree (pounds)

Row.	8	7	6	5	4	3	2	1	Total yield per row.
37	387	250	550	362	212	437	337	212	2,747
38	187	200	175	400	350	100	500	412	2,324
39	337	112	575	550	300	550	475	262	3,161
40	425	437	400	450	250	362	287	437	3,048
41	425	450	425	375	525	375	437	375	3,387
42	100	212	350	575	337	137	337	300	2,348
43	325	437	337	375	500	550	350	362	3,236
44	125	350	262	437	212	325	475	375	2,561
45	375	400	350	412	550	450	437	350	3,324
46	300	450	312	250	525	337	437	337	2,948
47	512	487	212	325	425	500	437	300	3,198
48	162	312	250	37	537	300	375	250	2,223
49	275	375	50	286	475	275	412	225	2,087
50	175	462	362	387	187	337	437	212	2,559
51	312	387	312	350	262	337	500	475	2,935
52	125	400	375	150	375	375	50	300	2,150
53	300	375	250	312	175	375	450	337	2,574
54	250	200	250	250	75	300	262	75	1,662
55	350	175	450	261	287	87	312	100	2,548
56	187	150	50	350	375	100	275	425	1,912
57	300	300	262	375	325	425	350	225	2,562
58	200	187	387	362	425	200	312	375	2,448
59	250	100	337	362	312	100	225	187	1,873
60	187	200	337	337	450	300	200	225	2,236
61	112	287	275	325	325	200	150	150	1,724
62	50	125	200	375	437	275	275	125	1,862
63	187	100	287	50	87	150	325	287	1,473
64	162	150	187	200	50	75	100	250	1,174
Total yield per row	7,082	8,070	8,469	9,520	9,345	8,334	9,519	7,945	68,284

ductivity, may have been instrumental in causing such a variation in yield.

TABLE I.—*Range of variability in crop production of fruit and nut trees*

[Extremes and range expressed as percentages of the mean yields of the respective plantations]

Kind of fruit.	Mean yield.	Extreme yield.	Range.	Mode.	Skewness.
	Pounds.	*Per cent.*	*Per cent.*	*Pounds.*	
Naval oranges (Arlington)	137±1.6	9.1 to 246.3	237.2	129.8	0.143±0.025
Naval oranges (Antelope Heights)	186±1.7	33.4 to 193.8	160.4	186.1	.001± .037
Valencia oranges	246±4.3	5.1 to 188.0	182.9	270.8	− .250± .053
Eureka lemons	270±2.9	32.4 to 180.3	147.9	299.9	− .359± .043
Seedling walnuts	86±1.6	14.4 to 276.1	261.7	75.6	.269± .049
Jonathan apples	304±5.6	12.3 to 193.2	180.9	345.1	− .329± .055

The biometrical constants for the several plantations are given in Table II. The oranges, lemons, and apples, as might be expected, show less variability than the seedling walnuts. The coefficient of variability of the clonal varieties ranges from 29.72 to 41.23 per cent. This total range of only 11.51 per cent shows a marked similarity of the extent of variation.

TABLE II.—*Variability in yield of the different individual fruit trees*

Kind of fruit.	Crop.	Acreage per tree.	Total number of trees.	Mean yield per tree.	Standard deviation.	Coefficient of variability.	Probable error.	
							Pounds per tree.	Percentage of mean.
				Pounds.	*Pounds.*			
Naval orange (Arlington)	1915	0.011	1,000	137.6±1.2	54.42±0.82	39.55±0.68	37	26.67
Naval orange (Antelope Heights)	1916	.011	495	186.2±1.7	55.33±1.19	29.72±0.69	37	20.05
Valencia orange	1915	.011	240	246.3±4.3	97.84±3.01	39.72±1.40	66	26.79
Eureka lemon	1915	.013	364	270.7±2.9	81.38±2.03	30.06±0.81	55	20.28
Seedling walnut	1915	.057	280	99.8±1.9	47.77±1.36	47.86±1.64	32	32.28
Seedling walnut	1916	.057	280	77.6±1.7	41.94±1.19	53.91±1.92	28	36.36
Seedling walnut, average	1915 and 1916	.057	280	86.4±1.6	40.10±1.14	46.41±1.58	27	31.30
Jonathan apple	1914	.011	224	303.9±5.6	125.30±4.00	41.23±1.52	85	27.81

The probable error, expressed in pounds of fruit per tree, is the greatest in case of the Jonathan apple, amounting to 85 pounds, while the Valencia orange and the Eureka lemon fall to 66 and 55 pounds, respectively. Such probable errors, expressed in pounds per tree, are not comparable, however, unless the mean yields are approximately the same. The probable error expressed as a percentage of the mean is therefore added to Table II to make it more easily compared with tables of other writers who have seen fit to use this constant rather than the coefficient of variability.

METHODS OF CALCULATING VARIABILITY

The yields of the various fruit plantations have been studied, with trees singly and combined into plots of various sizes. The coefficient of variability and probable error have been used as the basis of comparison in most cases.

Plots of different sizes necessarily have varying mean yields per plot; therefore the coefficient of variability is more readily interpreted than the standard deviation. The probable error may only be used with accuracy in cases where the number of variants is relatively large and their distributions normal.

VARIABILITY OF ORCHARD PLOT YIELDS

EFFECT OF INCREASING THE NUMBER OF ADJACENT TREES PER PLOT

The first point studied was the effect of increasing the number of adjacent trees per plot, measured by the coefficient of variability. Based on the theory of random sampling of variables, the average production of 10, or even 5, trees should be a more typical sample of the orchard than that

of 1 tree. The reduction of the coefficient of variability by combining a number of adjacent trees in a plot would, however, be expected to fall short of the theoretical reduction, because such a combination may have a tendency to group trees of similar productivity together. Gradual soil variation from one side of the plantation to the other, or irregularities of the field which are larger than the area taken up by a single tree, will tend to bring about a correlation between the yields of adjacent trees.

For practical purposes the two more or less antagonistic sources of variation between plots may be arranged in two groups:

1. Those which may cause the variations to become greater as the size of the plot increases—for example, variation in soil productivity.

2. Those which may cause the variations to become less as the size of the plot increases—for example, variations in inherent productiveness of the trees. This may depend to no small degree on the variation in vigor and character of growth of the rootstock. Measured by crop production, it may be practically impossible with grafted or budded trees to separate by mere inspection the variation which may be caused in inherent qualities of the bud from those of the rootstock on which it is propagated.

The reduction of the coefficient of variability in the several plantations as a result of increasing the number of adjacent trees per plot is shown in Table III. The acreage per plot is recorded for sake of comparison with similar work by agronomists, where the size of the plots studied has been dependent entirely on acreage rather than number of plants to the plot. Other biometrical constants are likewise included for ease of comparison with above-mentioned studies.

The effect of increasing the number of adjacent trees per plot on reducing the coefficient of variability between the plots of all the fruit crops studied is shown in the summary of Table III, and figure 7 shows the same thing graphically. The curves show a marked similarity between the varieties of fruits and agree quite closely in demonstrating that there is little to be gained in including more than eight adjacent trees in a plot. As a rule, there is a rapid reduction in the coefficient, as progress is made from a 1-tree to an 8-tree plot. Increasing the plot above eight adjacent trees shows only a comparatively small reduction of the coefficient of variability. In fact, the reduction is not significant when the probable errors[1] are considered. The Antelope navels and apples show a reduction slightly less than the probable error between a 4-tree and an 8-tree plot. Again, the lemons show an apparently exceptional reduction when the 16-tree plot is compared with the 24-tree plot. The same is true of the 8- and 16-tree plots of apple trees. These exceptions are in part explained, where they concern the larger plots,

[1] The probable error of the difference between two averages A_1 and A_2, of which the probable errors E_1 and E_2 are known, is the square root of the sum of the squared probable errors; or probable difference of $A_1 - A_2 = \pm\sqrt{E_1^2 + E_2^2}$

by the fact that in a given area, as the size of the plot increases the number of plots necessarily decreases, and thus lessens the reliability of the comparisons. Thus, these exceptions may be due to chance in a small population and might not hold true with a larger number of variants.

TABLE III.—*Effect of increasing the number of adjacent trees per plot*

Kind of tree.	Number of trees per plot.	Number of plots.	Acreage per plot.	Mean yield per plot.	Standard deviation.	Coefficient of variability.	Probable error.	
							Pounds per tree.	Percentage of mean.
				Pounds.	*Pounds.*			
Navel oranges (Arlington)	1	1,000	0.011	137.6± 1.2	54.42± 0.82	39.55±0.68	37	26.67
Do	2	500	.022	275.6± 2.9	95.60± 2.03	34.68±0.82	32	23.39
Do	4	250	.044	551.6± 7.4	173.89± 5.24	31.52±1.05	29	21.26
Do	8	125	.088	1,100.8± 19.2	319.00± 13.60	28.98±1.30	27	19.55
Do	16	60	.176	2,220.0± 56.0	642.60± 39.58	28.95±1.92	27	19.53
Do	24	40	.264	3,343.8± 92.5	867.70± 65.43	25.95±2.08	24	17.50
Navel oranges (Antelope Heights)	1	495	.011	186.2± 1.7	55.33± 1.19	29.72±0.69	37	20.05
Do	2	247	.022	373.0± 4.2	97.21± 2.95	26.06±0.84	33	17.58
Do	4	125	.044	742.7± 10.0	164.37± 7.07	22.13±1.00	28	14.93
Do	8	61	.088	1,460.7± 26.7	309.05± 18.90	21.16±1.35	26	14.27
Do	16	30	.176	2,973.3± 68.4	555.55± 48.38	18.68±1.68	23	12.60
Do	24	20	.264	4,450.0±121.6	806.54± 86.01	18.10±1.91	23	12.21
Valencia oranges	1	240	.011	246.3 ± 4.3	97.84± 3.01	39.72±1.40	66	26.79
Do	2	120	.022	487.92± 10.0	161.80± 7.04	33.16±1.61	55	22.37
Do	4	60	.044	991.7 ± 23.9	275.20± 16.92	27.75±1.84	46	18.72
Do	8	30	.088	1,966.7 ± 59.4	482.60± 42.02	24.54±2.26	41	16.55
Do	16	15	.176	3,813.3 ±158.6	910.58±112.13	23.88±3.10	38	16.11
Do	24	10	.264	5,880.0 ±243.0	1,140.00±171.90	19.39±3.03	32	13.08
Eureka lemons	1	364	.013	270.7 ± 2.9	81.38± 2.03	30.06±0.81	55	20.28
Do	2	182	.026	544.3 ± 6.8	135.40± 4.78	24.88±0.93	46	16.78
Do	4	91	.053	1,081.0 ± 17.3	245.35± 12.27	22.70±1.19	41	15.31
Do	8	42	.106	2,172.0 ± 46.7	448.50± 33.01	20.65±1.57	38	13.93
Do	16	21	.211	4,395.0 ±121.7	827.00± 86.07	18.82±2.02	35	12.69
Do	24	13	.317	6,692.0 ±153.5	820.30±108.50	12.26±1.65	23	8.27
Seedling walnuts	1	280	.057	86.4 ± 1.6	40.10± 1.14	46.41±1.58	27	31.30
Do	2	140	.114	178.7 ± 3.6	62.40± 2.52	34.92±1.57	21	23.55
Do	4	70	.228	353.6 ± 8.6	106.40± 6.06	30.09±1.86	18	20.30
Do	8	35	.456	717.1 ± 20.7	181.24± 14.60	25.27±2.16	16	17.04
Do	16	17	.912	1,409.4 ± 58.0	354.40± 41.00	25.15±3.07	15	16.96
Do	24	11	1.368	2,154.6 ±103.3	508.00± 73.05	23.58±3.53	14	15.90
Jonathan apples	1	224	.011	303.9 ± 5.6	125.30± 4.00	41.23±1.52	85	27.81
Do	2	112	.022	609.8 ± 12.3	193.00± 10.44	31.65±1.87	65	21.35
Do	4	56	.044	1,210.7 ± 28.8	319.40± 20.40	26.38±1.80	54	17.79
Do	8	28	.088	2,414.3 ± 76.9	603.38± 54.38	24.99±2.39	51	16.86
Do	16	14	.176	4,864.3 ±175.5	973.68±124.11	20.02±2.65	41	13.50
Do	24	9	.264	7,277.8 ±313.9	1,396.00±221.90	19.18±3.16	35	12.94

SUMMARY

Number of trees per plot.	Coefficient of variability.							Average reduction of coefficient of variability by increasing number of adjacent trees per plot.	
	Navel oranges.		Valencia oranges.	Eureka lemons.	Seedling walnuts.	Jonathan apples.	Average.		
	Arlington.	Antelope Heights.						Increase from—	Average reduction.
1	39.55±0.68	29.72±0.69	39.72±1.40	30.06±0.81	46.41±1.58	41.23±1.52	37.78±0.52		
2	34.68±0.82	26.06±0.84	33.16±1.61	24.88±0.93	34.92±1.57	31.65±1.87	30.89±0.55	1 to 2	6.89±0.76
4	31.52±1.05	22.13±1.00	27.75±1.84	22.70±1.19	30.09±1.86	26.38±1.80	26.76±0.62	2 to 4	4.13±0.83
8	28.98±1.30	21.16±1.35	24.54±2.26	20.65±1.57	25.27±2.16	24.99±2.39	24.27±0.77	4 to 8	2.49±0.99
16	28.95±1.92	18.68±1.68	23.88±3.10	18.82±2.02	25.15±3.07	20.02±2.65	22.58±1.01	8 to 16	1.69±1.27
24	25.95±2.08	18.10±1.91	19.39±3.03	12.26±1.65	23.58±3.53	19.18±3.16	19.74±1.08	16 to 24	2.84±1.48

The averages of all six fruit plantations show that there is a rapid reduction of the coefficient of variability until the 8-tree plot is reached, but from then on the reduction is less in comparison with the probable error. (See Table III, summary, and figure 8.)

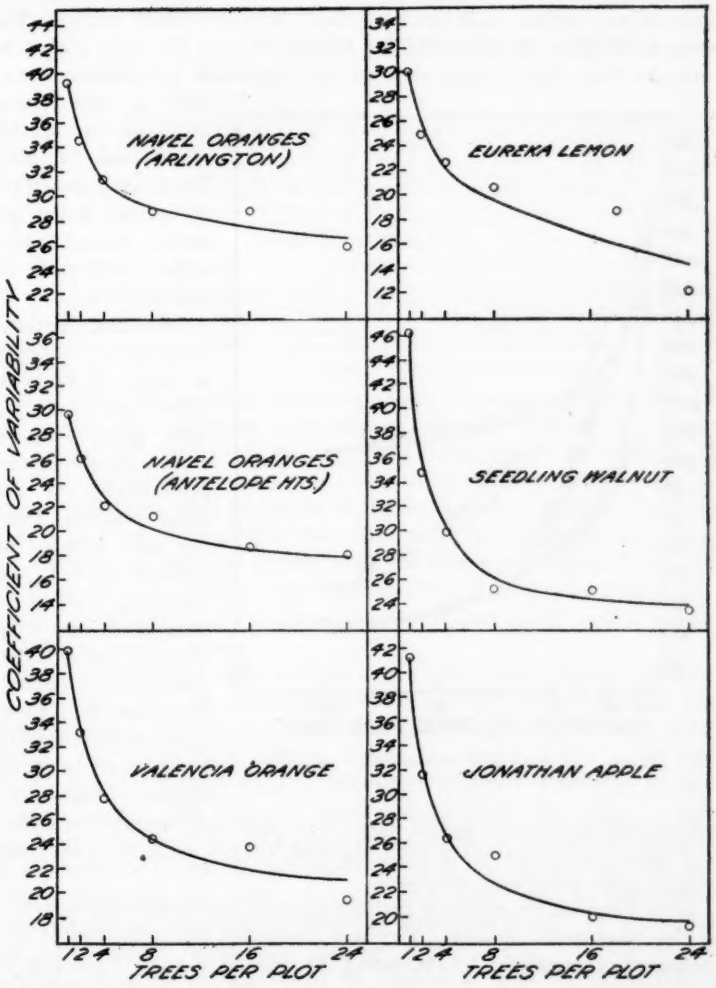

FIG. 7.—Graphs of the reduction of the coefficient of variability by increasing the number of adjacent trees to the plot.

Although the average reduction of the coefficient between the 8-and 16-tree plots is doubtful when compared with the probable error, this reduction in all six cases is constant—that is, the variation occurs in

one direction only, and therefore has more significance than is indicated by a mere comparison of the averages.

EFFECT OF SYSTEMATIC DISTRIBUTION OF PLOTS OVER THE AREA STUDIED

The importance of distributing plots over the experimental area is more or less obvious, and has been dwelt upon by many writers. Its value arises from the fact that the soil varies over the area, and it is better to have similar-sized plots on both high- and low-yielding areas than to have them solely on one or the other kind of soil. The method should be of special value on areas which vary rather uniformly in one direction.

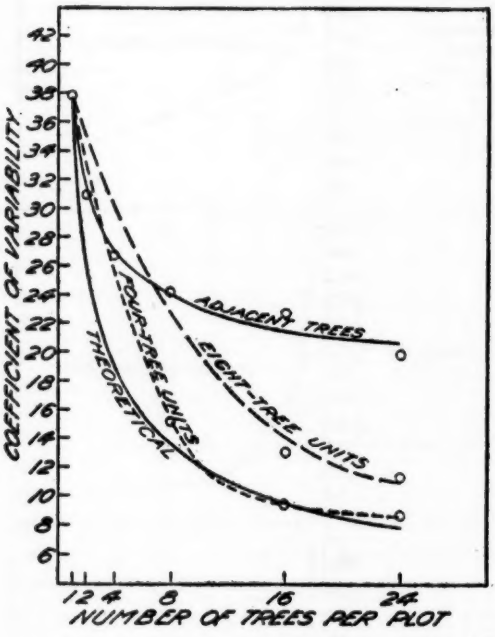

FIG. 8.—Graphs of the reduction of the coefficient of variability by increasing the number of trees to the plot.

Increasing the number of trees to the plot in scattered units of either four or eight trees gives a more typical sample of the productivity of the total planting than the same number of adjacent trees. In scattering the plots throughout the area studied, they were systematically repeated. For example, if there were 100 plots in all to be grouped in pairs, the first and fifty-first, and the second and fifty-second were united, and so on through the series. If a quadruple series was desired, the first, twenty-sixth, fifty-first, and seventy-sixth plots were combined.

Table IV shows the results of scattering 4- and 8-tree plots, respectively, in the plantations studied. Figure 8 illustrates the reduction of the coefficient of variability by increasing the number of trees to the plot in both 4- and 8-tree scattered units, compared with the average coefficient of variability for the several fruits by increasing the size of a plot from 1 to 24 adjacent trees, together with the theoretical curve calculated from the mean coefficient of variability of all the 1-tree units.

A comparison of the curve for adjacent trees and those for scattered units shows at once the marked decrease in favor of the scattered units.

TABLE IV.—*Effect of increasing number of trees per plot in scattered units*

FOUR TREES IN A UNIT

Unit No.	Total number of trees per plot.	Coefficient of variability.						
		Navel oranges.		Valencia oranges.	Eureka lemons.	Seedling walnuts.	Jonathan apples.	Average.
		Arlington.	Antelope Heights.					
1....	4	31.52±1.00	22.13±1.00	27.75±1.80	22.70±1.10	30.09±1.85	26.38±1.80	26.76±0.60
2....	8	18.59±0.82	13.06±0.85	16.04±1.43	14.76±1.07	12.36±1.01	15.89±1.47	15.12±0.47
3....	12	15.11±0.81	11.07±0.88	16.48±1.81	12.61±1.16	14.17±1.41	12.04±1.37	13.58±0.53
4....	16	10.76±0.66	9.52±0.87	10.39±1.29	8.01±0.81	8.59±0.99	8.44±1.08	9.29±0.40
5....	20	11.15±0.76	5.74±0.58	9.28±1.28	9.48±1.07	6.97±0.89	7.75±1.13	8.40±0.40
6....	24	9.89±0.74	6.38±0.72	13.16±2.02	5.38±0.66	10.05±1.45	6.12±0.97	8.49±0.49

EIGHT TREES IN A UNIT

1....	8	28.98±1.30	21.16±1.35	24.54±2.26	20.65±1.57	25.27±2.16	24.99±2.39	24.27±0.77
2....	16	17.49±1.09	9.21±0.83	15.22±1.92	10.98±1.17	10.94±1.27	13.20±1.69	12.84±0.56
3....	24	11.78±0.89	7.44±0.84	14.34±2.21	11.70±1.44	11.35±1.63	10.99±1.75	11.27±0.63
4....	32	14.85±1.30	8.87±1.14	8.72±1.57	7.56±1.16	6.68±1.13	10.54±1.90	9.54±0.57
5....	40	10.09±0.96	8.60±1.24	5.24±1.02	7.81±1.32	11.01±2.01	4.94±1.05	7.95±0.54

The 4-tree unit apparently gives a greater degree of accuracy than the 8-tree unit with the same total number of trees. This point is clearly shown by the curves. In considering a total number of either 8, 16, or 24 trees the curve for the 4-tree unit approaches more nearly the theoretical curve than the curve of the 8-tree unit. With a total number of 24 trees, for example, the 4-tree units would be scattered about regularly in six different places, while the 8-tree units would be located in three different places. The larger number of unit plots thus gives a more typical sample of the area than half as many units with twice the number of trees in a unit.

In combining both the 4- and the 8-tree units in the regular scattering of ultimate plots, an attempt was made to throw both high- and low-yielding small plots into a combination, although a systematic distribution was maintained. The fact that the curve of the 4-tree units drops below the theoretical in one place indicates that this attempt was successful. An ordinary regular scattering of the ultimate plots might not approach the theoretical curve of random sampling so closely if a knowledge of the relative productivity of the soil was not available before arranging the plots. A 4-tree unit might not be practical in tests of cultural methods, fertilizer, or irrigation trials. In such trials an 8-tree plot is usually the smallest practical unit. In the case of walnuts, however, which should be planted at least 50 by 50 feet, a 4-tree unit in a single row would allow for a space 50 feet wide by 200 feet long, and, if guard rows were planted between the experimental trees, the plots would be 100 by 250 feet, a very practical size upon which to handle orchard machinery. For the trial of rootstocks, pruning experiments, variety tests, etc., the 4-tree plot is a practical-sized

unit and could be expected to give more reliable results if repeated at four regularly-placed intervals than either two 8-tree units, or 16 adjacent trees—that is, such a regular scattering of the several units which make up the combination plot reduces the error of the final comparisons which is caused by the variation in soil productivity.

The fact that marked soil variations occur which tend to make adjacent trees or adjacent plots yield alike, even on soils which were chosen because of their apparent uniformity, is well shown by the work of Harris (*1915*). The criterion for the measurement of such variability proposed by this author is the coefficient of correlation between neighboring plots of the field.[1] Applying this to the Arlington navel oranges, the writers have calculated the correlation between the yield of the 8-tree plot as the ultimate unit, and the yield of the combination of four such adjacent plots and it was found that

$$r = +0.533 \pm 0.085.$$

This shows a marked correlation, indicating a pronounced heterogeneity in the soil of this grove, influencing fruit production.

However, when we calculate the correlation between the 8-tree plot as the ultimate unit and the yield of the combination of four such systematically scattered plots, it is found that—

$$r = +0.137 \pm 0.120$$

This coefficient is practically equal to its probable error and can be regarded as significantly zero. This is merely another means of calculating the value of scattering a 32-tree plot in four ultimate plots of 8 trees each rather than selecting 32 adjacent trees.

DEGREE OF ACCURACY EXPECTED WITH A PLOT OF A GIVEN SIZE

Assuming, for example, that experimental plots have been laid out in the navel oranges (Arlington) with a total of 32 trees to the plot in four scattered units of eight trees each, the question might logically be asked, "What differences in the yields of such plots can safely be attributed to differential treatment as different methods of irrigation or fertilization, and what may probably be due to mere chance because of soil heterogeneity and the fluctuating variation of the trees?"

Table IV shows a coefficient of variability of 14.85 ± 1.30 in this plantation laid out in 32-tree plots of four scattered units of eight trees each. The probable error,[2] then, in this example, that such a plot of 32 trees is typical of the area in question, is $14.85 \times 0.6745 = \pm 10.02$

[1] The formula used is

$$r_{p_1 p_2} = \frac{\{[S(C^2 p) - S(p^2)]/m[n(n-1)]\} - \bar{p}^2}{s p^2}$$

where p = yield of an individual plot; m = number of larger plots, each made up of n contiguous ultimate units; C_p = yield of the larger combination plots; S = summation of the yields of all the ultimate or combination plots of the field.

[2] The probable error of a single variant of a population may be defined as that departure from the mean on either side, within which exactly one-half of the variants are found. Expressed as a percentage of the mean, it is determined by multiplying the coefficient of variability by 0.6745.

per cent of the mean production. That is, the chances are even that any plot as described, of 32 trees, will fall within ±10.02 per cent above or below the true mean. The chances are equally as good that such a plot will not fall within the accuracy of ±10.02 per cent of the mean. In comparing two such plots, both with the same probable error of ±10.02 per cent, the probable error of such a comparison will be greater than the probable error of one—that is, it will be equal to ±10.02 per cent $\times\sqrt{2}=\pm 14.17$ per cent. Therefore, if plots undergoing differential treatment vary from each other by only ±14.17 per cent of the mean of the plantation, half the time such differences in yield may be due to the treatment, and half the time they may be due to casual variation.

It is clear, then, if fertilizer or irrigation experiments laid out in such plots differ from each other by only 14.17 per cent of the mean production of the total area, we are not assured beyond an even chance that the difference is a real difference due to the factors which are being experimented upon. So slight an assurance can hardly be expected to be sufficiently reliable to prompt a farmer to purchase fertilizer, to change his method of irrigation, or to undertake any new business; much less will this assurance justify an experimenter in drawing conclusions from the result of a field trial.

Our present knowledge of orchard fertilization in the arid West will hardly justify any assumption on our part more reliable than an even chance that one fertilizer will produce an increased yield of fruit compared with another, or even cause an increase over an untreated plot. The same thing may be said in comparing different methods of irrigation. In most cultural trials we would therefore be comparing two results where the difference may occur in either direction. (Tables V and VI.)[1]

TABLE V.—*Table of odds for differences which may occur in either direction*

Difference from the mean in terms of probable error.	Difference between two results in terms of probable error of each result.	Odds against such difference occurring under uniform conditions.
1.00	1.41	1 to 1
1.25	1.76	3 to 2
1.44	2.03	2 to 1
1.71	2.41	3 to 1
1.90	2.68	4 to 1
2.00	2.83	9 to 2
2.05	2.87	5 to 1
2.50	3.53	10 to 1
2.93	4.13	20 to 1
3.00	4.24	22 to 1
3.20	4.51	30 to 1
4.00	5.66	140 to 1
4.90	6.93	1,000 to 1
5.00	7.07	1,350 to 1

[1] Tables V and VI are taken from the writings of Wood (*1911*) who in turn adopted them "from one of the standard reference books on astronomy."

TABLE VI.—*Table of odds for differences which may occur in one direction only*

Difference from the mean in terms of probable error.	Difference between two results in terms of probable error of each result.	Odds against such difference occurring under uniform conditions.
1.00	1.41	3 to 1
1.25	1.76	4 to 1
1.44	2.03	5 to 1
1.58	2.23	6 to 1
1.71	2.41	7 to 1
1.81	2.55	8 to 1
1.90	2.68	9 to 1
2.00	2.83	10 to 1
2.48	3.50	20 to 1
2.70	3.81	30 to 1
2.89	4.07	40 to 1
3.00	4.24	44 to 1
3.03	4.28	50 to 1
3.44	4.85	100 to 1
4.00	5.66	290 to 1
5.00	7.07	2,700 to 1

On assuming that a 10-to-1 chance is a reasonable assurance, the question logically arises, "What difference between any two plots must be manifested for this degree of confidence that the difference is due to treatments applied?" By referring to columns 2 and 3, Table V, one may find the difference in terms of the probable error which is necessary between two results to obtain this degree of reliability. Here it can be seen in column 2 that there must be a difference 3.53 times the probable error to give the odds of 10 to 1 in column 3 against such a difference occurring under uniform conditions. Thus we find in this example that the difference between two 32-tree plots in four scattered units must be at least 14.17 per cent $\times$ 3.53 = 50.02 per cent of the mean production, to give the assurance of a 10-to-1 chance that the difference is due to fertilizer, irrigation, or whatever factors are under consideration.[1] Even with this difference, conclusions based on such results obtained in this navel orange (Arlington) grove may be correct 10 times out of 11 and wrong once out of 11 times.

On turning now to the 32-tree plot of adjacent trees with navel oranges (Arlington), it is seen the probable error is 16.42 per cent. To proceed as before, 16.42 per cent, the probable error of one plot, $\times \sqrt{2} =$ 23.22 per cent, the probable error of the difference; $23.22 \times 3.53 = 81.97$ per cent. Therefore a difference between two such plots of 81.97 per cent of the mean of the total area would be necessary to give the assurance that such differences are real 10 times out of 11 and due to pure chance

[1] If comparisons were being made between two radically different treatments which were known to produce different effects in fruit production, such as irrigation compared with dry farming, or the use of large quantities of stable manure on light soils, compared with no manuring, then reference should have been made to columns 2 and 3 in Table VI, which are based on the differences occurring in one direction only.

only once out of 11 times; yet this grove was chosen because of its apparent regularity, for it has been judged sufficiently uniform for plot trials.

The point might be justly raised that the small number of plots involved in the above calculations are not sufficient to give the laws of chance a fair opportunity of asserting themselves. On laying the area out in plots of adjacent trees there were 30 plots, while made up of scattered units there were 31 plots. Figure 9 shows the distribution of these plots, together with the theoretical curve, which was caclulated for the scattered-unit curve. The scattered-unit curve closely approaches the theoretical normal curve of errors, and therefore reliance can be placed upon its probable error.

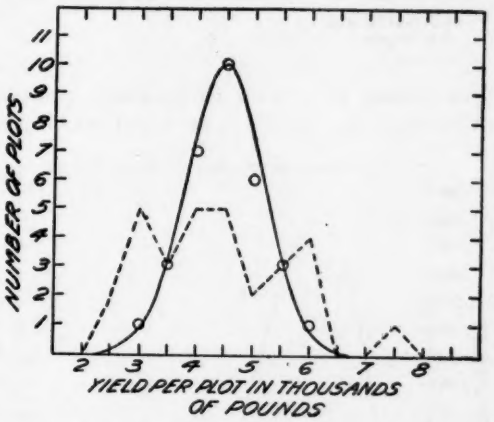

FIG. 9.—Graphs of production, 32-tree plot, navel oranges (Arlington).
——— Scattered in four 8-tree units.
- - - - Adjacent 32-tree units.

In the case of the plots of adjacent trees, however, the 30 units are not sufficient to give the laws of chance fair play. Table VII sums up the results of the foregoing calculations, adding extreme and mean yields of the two different types of plots and the theoretical probable error. The theoretical probable error based on the theory of random sampling for a hypothetical 32-tree plot is the probable error of one tree $26.67 \div \sqrt{32} = 4.71$ per cent. This is readily calculated from the distribution of the yields on a one-tree unit, the curve of which is shown by figure 10. The large number of trees involved, even though the distribution is not normal, justifies the use of the probable error as a minimum probable error. Based upon the theory of random sampling, two hypothetical 32-tree plots with a probable error of 4.71 per cent should show a minimum difference of $(4.71 \times \sqrt{2} \times 3.53) = 23.51$ per cent to give an assurance of a 10-to-1 chance that such a difference is real and not due to casual variation. Therefore, if the calculations in Table VII based on adjacent trees can not be fully relied upon because of the small number (30) in the population and because their distribution is not normal, we may at least reasonably expect that the necessary difference between two such plots will fall between the theoretical 23.51 per cent and 81.97 with a practical certainty that it will be greater than 50.02 per cent of the mean.

TABLE VII.—*Comparison of the reliability of a plot of 32 adjacent trees with that of a 32-tree plot of four scattered units of 8 trees each. Navel oranges (Arlington)*

Plot.	Extreme yields per plot.	Mean yield per plot.	Standard deviation.	Probable error per plot (percentage of mean).	Difference necessary to give 10-to-1 assurance.	
					Percentage of the mean.	Observed yield.
	Pounds.	*Pounds.*	*Pounds.*			*Pounds.*
Adjacent trees	2,500 to 7,500	4,367±131	1,063±93	±16.42	±81.97	±3,580
Scattered-unit plot	3,000 to 6,000	4,484± 81	666±57	±10.02	±50.02	±2,243
Theoretical (based on random sampling)				±4.71	±23.51	±1,035

On turning now to the navel oranges (Antelope Heights), which the calculations show to be the most uniform planting of any observed, the question might arise as to what degree of reliability may be expected in comparing two 16-tree plots. Table VIII shows a comparison between such plots made up of adjacent trees and plots composed of scattered units of four trees each together with the theoretical. A much greater range is found with the plot of adjacent trees, while the necessary difference between two plots for a 10-to-1 assurance is 62.94 per cent. With plots made up of scattered units the necessary difference is 32.05 per cent. The heterogeneity of this soil is clearly shown by both the increase in range and the increased probable error when plots are composed of adjacent trees. This block of trees, however, appeals to the observer as unusually uniform and would be considered desirable for plot experimentation, the fluctuation in the productivity of the trees approaching closely

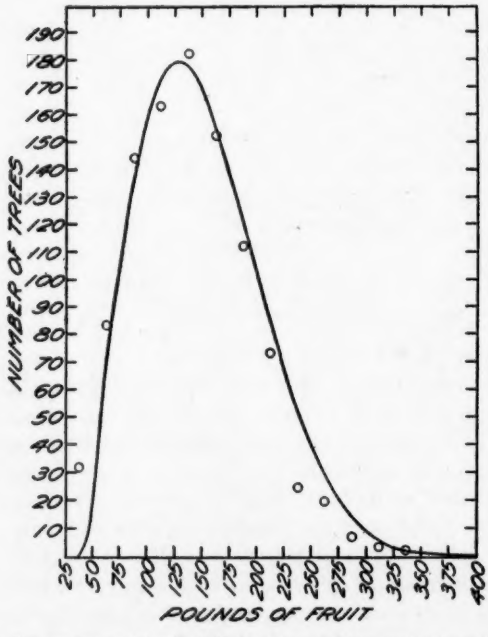

FIG. 10.—Curve of yields of individual trees, navel orange (Arlington).

the normal curve (see fig. 11); the mean production per tree is 186.2 ± 1.7 pounds, standard deviation 55.33 ± 1.19, coefficient of variability 29.72 ± 0.69 per cent, probable error 20.05 per cent of the mean, skewness 0.001 ± 0.037. Nevertheless, if devoted to plot experiments, a difference between two plots of 16 adjacent trees each, of even 62.94 per cent of the mean production, might be due to differential treatment 10 times out of 11 and due to casual variations of soil and trees once out of 11. The calculations sound a note of warning against drawing conclusions between such plots if the differences are less than 50 per cent of the mean production of the plantation, provided we wish to have such conclusions as dependable as a 10-to-1 chance.

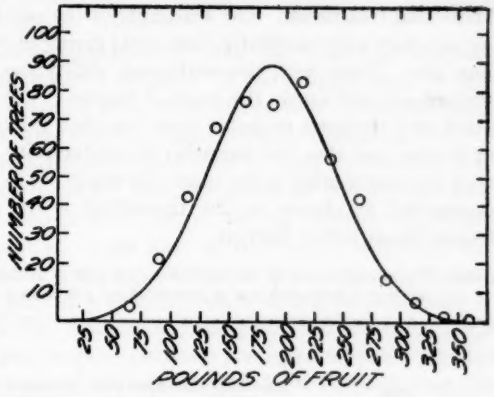

FIG. 11.—Curve of yield of individual trees, navel orange (Antelope Heights).

TABLE VIII.—*Comparison of the reliability of a plot of 16 adjacent trees with that of a plot of 16 trees of four scattered units of 4 trees each. Navel oranges (Antelope Heights)*

Plot.	Extreme yields per plot.	Mean yield per plot.	Standard deviation.	Probable error per plot (percentage of mean).	Difference necessary to give 10-to-1 assurance.	
					Percentage of the mean.	Observed yield.
	Pounds.	*Pounds.*	*Pounds.*			*Pounds.*
Adjacent trees	1,800 to 4,000	2,973±68	556±48	±12.61	±62.94	±1,871
Scattered unit plot	2,400 to 3,600	2,960±36	282±25	± 6.42	±32.05	± 949
Theoretical				± 5.01	±25.03	± 746

An example taken from the Jonathan apple orchard may well be considered. Suppose it is desired to know the necessary difference which must exist between plots of 16 trees each to give us the reliance of a 10-to-1 chance that it is due to differential treatment (a) when the plots are made up of adjacent trees, (b) when the plots are made up of two scattered units of eight trees each, and (c) when the plots are made up of four scattered units of four trees each.

The probable errors in the above cases, a, b, and c, are ±13.49, ±8.90, and ±5.69 per cent, respectively. By proceeding as before it is found that the necessary difference for a 10-to-1 chance is as follows: (a)

±67.35 per cent; (b) ±44.44 per cent; (c) ±28.42 per cent. Table IX summarizes the results. It seems probable, therefore, that a difference between two 16 adjacent tree plots of less than 50 per cent of the mean production should be considered with caution before attributing it to differential treatment. The scattering of the units of the plots increased the accuracy very decidedly, four units giving more accurate comparison than two. Even with this scattering, differences of less than 30 or 40 per cent are well within the realm of chance. The apparent cause for the 4-unit plot having a probable error less than the theoretical is accounted for by the fact that the variation in productivity of the soil was known when the distribution of the units was made. The results might not have approached so closely to the theoretical if the distribution had been decided upon before harvest.

TABLE IX.—*Comparison of the reliability of a plot of 16 adjacent trees with that of two units of 8 trees each and of four units of 4 trees each. Jonathan apples*

Plot.	Extreme yields per plot.	Mean yield per plot.	Standard deviation.	Probable error per plot (percentage of mean).	Difference necessary to give 10-to-1 assurance.	
					Percentage of the mean.	Observed yield.
	Pounds.	*Pounds.*	*Pounds.*			*Pounds.*
(a) 16 trees adjacent	3,000 to 6,500	4,864±176	974±124	±13.49	±67.35	±3,276
(b) 2 units of 8 trees	3,900 to 6,100	4,857±115	641± 82	± 8.90	±44.44	±2,158
(c) 4 units of 4 trees	3,800 to 5,400	4,871± 74	411± 52	± 5.69	±28.42	±1,384
Theoretical (based on random sampling)				± 6.95	±34.70	±1,687

The large probable errors which are apparently always present with plot trials of fruit trees emphasize the importance and value of obtaining individual tree records of experimental orchards before differential treatment is started. The probable errors will likely differ somewhat from year to year, and possibly be further influenced by the advanced age of the trees. Nevertheless, if but one or two years' records of mature trees are available before differential treatment is commenced, at least some idea can be obtained of the casual variation of the plots—that is, limitations can be placed beyond which observed differences in plots may be due to chance rather than to the factors under experimentation. In the absence of such previous records, the employment of frequent controls or standard treatments may be indicative of the probable error of the entire area.

RELATION OF THE SHAPE OF THE PLOT TO THE VARIABILITY OF THE COMPARATIVE YIELDS

The shape of small plots may be of great importance when cultural operations are considered.

Lyon (*1912*) found no satisfactory evidence that long and narrow plots are less likely to error than square plots when no control plots are

used, but when controls are placed every second or third plot in the series, the evidence is in favor of the long and narrow plots. The use of every second or third plot in the series as a control plot obtains greater accuracy than when no controls are used and the average of the field is considered the normal yield for all plots.

A long plot is much more economical of time and labor than a square plot containing the same number of trees. If the orchard must be irrigated, a square plot containing nine trees will require three standpipes instead of one and also three times as many irrigation furrows.

Arguments have been advanced from time to time in favor of both the linear and the square plot. The advocates of the former have urged its reliability on two points: first, if the soil or other conditions change in a direction parallel to the plot, it will contain both high- and low-yielding areas, and the average will correct one or the other error; second, if conditions change in a direction more or less perpendicular to the plots, each plot will vary from its adjoining plot because of its shape by such a minimum that intercomparisons are more reliable. On the other hand, the advocates of the square plot have claimed that the arguments apply equally well to their case, provided the plot be made small enough.

In view of the importance of the shape of the plot for cultural operations, the writers have investigated the variability of plots of various sizes and shapes in three of the experimental orchards. The results (Table X) are for unrepeated plots and for one year's yield of fruit, except the apples, which are for two years. Considerable interest lies in the computation on the 9-tree plot in the total navel-orange grove, because it is based on a large population (1,000 trees) and because the soil is known to vary in a general way from northeast to southwest.

Comparisons were made between square plots containing 9 trees and linear plots consisting of a single row of 9 trees extending in the north-south and in the east-west direction. It is interesting to see how closely the coefficients of variability coincide in the three cases. The differences between the coefficients are only a fraction of the probable errors. Therefore on this lot of 1,000 orange trees there is no difference between a square and a linear plot of 9 trees, so far as the reliability of comparative yields is concerned. (The deviations were taken from the mean of all the plots of the grove.)

Similar comparisons were made on a selected block of 256 trees in this same grove. The coefficient of variability of the trees taken singly is 34.47 ± 1.14, which indicates that these trees were a fairly typical sample of the entire grove. The block was divided into plots of various shapes containing 16 trees. With one exception there is little difference in the variability of the plots, regardless of their shape. This exception is found in the linear plot of 1 by 16 trees extending from east to west, which has a lower coefficient than any other arrangement. In the block

of trees chosen there is a gradual variation of soil in this direction, and these plots therefore include both high- and low-yielding trees in about equal proportions, the difference from plot to plot being small, as shown in Table X.

TABLE X.—*Effect on variability of changing the shape of plots*

Kind of trees.	Number of trees per plot.	Shape of plots.	Coefficient of variability.
Total navel-orange grove (Arlington)	9	3 by 3	28.18±1.48
Do	9	1 by 9 N.-S.	28.92±1.46
Do	9	1 by 9 E.-W.	28.36±1.43
Portion of navel-orange grove (Arlington), 256 trees	1	1 by 1	34.47±1.14
Do	16	4 by 4	16.77±2.05
Do	16	2 by 8 N.-S.	17.17±2.10
Do	16	2 by 8 E.-W.	15.39±1.88
Do	16	1 by 16 N.-S.	16.36±2.00
Do	16	1 by 16 E.-W.	10.09±1.31
Walnuts, 1915	4	2 by 2	31.09±1.93
Do	4	1 by 4	30.40±1.89
Do	9	3 by 3	23.94±2.32
Do	9	1 by 9 N.-S.	26.23±2.57
Do	9	1 by 9 E.-W.	22.46±2.16
Do	24	4 by 6	21.62±3.25
Do	24	1 by 24	6.56±1.00
Apples, 1914–15	8	2 by 4	19.32±1.81
Do	8	1 by 8 N.-S.	18.70±1.74
Do	8	1 by 8 E.-W.	21.35±2.18
Do	16	4 by 4	15.13±1.97
Do	16	2 by 8 N.-S.	16.41±2.15
Do	24	4 by 6	13.75±2.36
Do	24	1 by 24	12.81±2.20

Changing the shape of plots of walnut trees had little effect upon the coefficient of variability except in the case of the larger plots. The coefficients vary only slightly from one another, regardless of shape, in the case of the 4- and 9-tree plots. In the 24-tree plot, however, there is a great difference between the 4- by 6- tree plot and the 1- by 24- tree plot in favor of the latter. It should be borne in mind, nevertheless, that a plot of 24 walnut trees is an abnormally large plot, and in the example just referred to it is a plot 250 feet by 350 feet, compared with one 100 feet by 1,250 feet. This may take into consideration marked variation in productivity, even on apparently uniform soil.

In the case of the apple trees there is little difference between the variability of plots of equal size but different shapes, even in the case of the 24-tree plots. Regarding the question of the shape of the plot, it therefore appears that in most cases there is no difference in the variability of a linear or a square plot.

proportional to their distance from the plot. This would obviously be

In the case of a large number of trees, where a linear plot is long enough to include both high- and low-yielding areas in each plot, there appears to be an advantage in favor of the linear plot. The great advantage of the linear plot in most cases is the economy of cultural operations. Another advantage will be discussed in the following section on the interpolation of control plots.

A carefully conducted fertilizer experiment with orchard trees requires a guard row between adjacent plots in order to eliminate the possibility of the fertilizer's affecting the margin of the next plot, since there are many observations which show that tree roots extend considerable distances and often reach the lines of adjoining rows (*Hedrick, 1914; Ballantyne, 1916*). It is obvious that it is more reliable to employ guard rows rather than to divide cultural treatments midway between tree rows. The number of guard trees required for square plots is smaller than for a linear plot containing the same number of trees. A 9-tree plot in the form of a square requires 7 guard trees; in the form of a linear plot it requires 11 guard trees. In the former case seven-sixteenths of all trees are in the guard rows, in the latter eleven-twentieths. As the size of the plot increases, the difference becomes greater. It thus becomes a question of the extent to which one is willing to go in enlarging the size of the linear plot before the increase in the number of trees in guard rows offsets the economy in cultural operations thus obtained.

USE OF CONTROL PLOTS

INTERPOLATED CONTROL PLOT

Agronomists are in the habit of using every third, fourth, or fifth row or plot in the experimental tract as a standard from which the normal yield of any intervening treated plot may be calculated. The nature of the crops and the cultural methods used commend this system of arrangement.

This method, like others, has its advantages and disadvantages. After the results are obtained there is still a need for a proper method of comparison. There are several different methods of estimating the "normal" yield of any plot. The "normal" (N) may be estimated by the formalu

$$\frac{C_1+C_2+\ldots.C_n}{n}$$

which is simply the mean of all the control plots in the area. If the soil of the area were uniform and all variations in the yield of the controls were purely chance variations, this method would give a satisfactory result. Again N may be estimated from the yields of the two nearest control plots. For example, if every third plot is a control and the arrangement is C_1, A, B, C_2, and so on, the normal for A would be $\frac{2}{3}C_1+\frac{1}{3}C_2$. In this way the yields of the controls receive weights inversely proportional to their distance from the plot. This would ob-

viously be satisfactory, provided there was no difference in the amount of variation between the plants on different plots and that the soil varied uniformly in one direction.

The first method mentioned computes the "normal" (N) for the whole area; the second, for a locus on that area. These normals may be combined to represent the resultant of both general and local conditions. Thus, the formula $N=\frac{1}{2}\ (C+\frac{2}{3}C_1+\frac{1}{3}C_2)$ indicates that N is the mean of the values for N computed by the two preceding formulas. This assumes an equal value for the adjacent control plots and the mean of all control plots. Its use as N brings up the calculated yields of plots on low-yielding areas and reduces the same on high-yielding areas. In the case of cereals there is usually small chance for difference in the yielding powers of a plot and its nearest control on account of their proximity; but in the case of orchard trees situated some distance apart there may be greater soil changes between adjacent plots, and consequently a marked difference in yield, aside from the effect of treatment, between a plot and its nearest control plots. The introduction of the mean of all control plots might be expected to introduce a stabilizing factor. In the formula $\frac{1}{2}\ (C+\frac{2}{3}C_1+\frac{1}{3}C_2)$ the mean of all control plots has equal weight with the normal derived from the nearest controls. Since the soil over any but very small areas may not be uniformly variable, it might seem more logical to weight the normal derived from the nearest controls more heavily than that derived from the mean of all control plots, and to combine the two. This has been done by Olmstead (*1914*) and others, making the formula $\frac{p_1C+p_2(\frac{2}{3}C_1+\frac{1}{3}C_2)}{p_1+p_2}$ in which p_1 and p_2 are constants arbitrarily chosen. Stockberger (*1916*) found satisfactory results by assigning the values $p_1=1$ and $p_2=3$.

The method used by the Office of Cereal Investigations, of the Bureau of Plant Industry, is $\frac{1}{2}(c+C_1)$, which employs half the sum of the mean of all control plots and the yield of the nearest control plot as the normal for any given plot.

Stockberger (*1916*) compared the relative precision of these formulas in computing the normal yields of plots of hops. Using the yields of six years he obtained the greatest precision from the formula

$$\frac{p_1C+p_2(\frac{2}{3}C_1+\frac{1}{3}C_2)}{p_1+p_2}$$

though no formula maintained the same relative rank throughout the six years. It would appear that there is no way of determining in advance the formula best suited to any particular case, at least not until more applications of the different formulas have been studied.

The five formulas above stated have been tested on the Arlington grove of navel oranges. The grove was parceled into linear plots of 10 trees each. Each alternate plot was designated as a guard row and

discarded from the calculation. This left 32 "treated" plots whose "normal" yields were computed. The arrangement of these plots and their yields are given in Table XI.

TABLE XI.—*Arrangement and yields of 10-tree plots of the navel-orange grove (Arlington)*

[Yield expressed in tens of pounds]

80 control	116*
105*	152 control
88H	184*
115*	155A
117G	164*
94*	154B
109 control	160*
118*	163 control
94F	147*
76*	122C
83E	142*
97*	137D
104 control	111*
101*	131 control
99D	148*
117*	154E
100C	165*
112*	134F
74 control	111*
101*	114 control
112B	109*
107*	123G
98A	78*
108*	97H
80 control	80*
108*	106 control
108H	137*
110*	135A
108G	84*
150*	88B
112 control	119*
124*	146 control
121F	151*
159*	174C
142E	132*
148*	182D
139 control	166*
153*	174 control
177D	180*
179*	169E
176C	131*
188*	162F
183 control	183*
178*	147 control
203B	188*
182*	168G
206A	223*
199*	201H
219 control	191*
191*	228 control

Computing the normal yields of these plots by the five formulas above described gives the results shown in Table XII.

TABLE XII.—*Value of different formulas for computing the coefficient of variability and probable error of yields of oranges in 10-tree plots in Arlington grove*

Formula used.	Standard deviation.	Coefficient of variability.	Probable error (percentage of mean).
	Pounds.		
1. $\frac{C_1+C_2.....+C_n}{n}$	363±31	26.55±2.39	17.91
2. $\frac{2}{3}C_1+\frac{1}{3}C_2$	171±14	12.52±1.07	8.44
3. $\frac{1}{2}(C+\frac{2}{3}C_1+\frac{1}{3}C_2)$	219±18	16.01±1.38	10.80
4. $\frac{p_1C+p_2(\frac{2}{3}C_1+\frac{1}{3}C_2)}{p_1+p_2}$	180±15	12.93±1.11	9.00
5. $\frac{1}{2}(C+C_1)$	233±20	16.78±1.46	11.32

*Guard row.

These figures show that there is less error in this case by the use of formulas (2) and (4) and that there was very little difference between these two.

This question of a "normal" yield of a plot depends obviously upon the portion of the population chosen as a standard and upon the method of calculation. Investigators differ in the choice of both of these factors.

An illustration may be given to show the results of calculating the normal from different standards. The coefficient of variability of the 10-tree plot of Arlington navel oranges was computed, taking the deviations from (a) the mean of all plots, (b) the mean of all control plots, and (c) the "normal" calculated by formula 2. The results are shown in Table XIII.

TABLE XIII.—*Deviation of yields from mean of area compared with deviation from 10-tree linear control plot*

Formula No.	Deviation.	Coefficient of variability.
1	Deviation taken from mean of all plots	26.00±2.30
2	Deviation taken from mean of control plots	26.55±2.39
3	Deviation taken from normal calculated from nearest control $\frac{2}{3}C_1+\frac{1}{3}C_2$	12.5±1.10

Since no differential treatments had been given these plots, the mean of all plots differs little from the mean of all control plots, the respective means being 1,376 and 1,367 pounds. The variability of plots calculated from these two standards is not significantly different. In the case of the deviation taken from the normal yield, however, there is a very significant decrease in variability, since there are discontinuous soil variations not recognizable in the general average of the area which are taken into account by this formula.

CHANCE ARRANGEMENT OF CONTROL PLOTS

A weakness of this system of comparison with interpolated plots lies in the possibility that the series of control plots may not be representative of the area. The plots chosen for controls may be on soil superior or inferior to that of the intervening plots. There are indications that this possibility may be more real than one would expect from purely random sampling.

A few computations will show the extent to which the different methods of choosing control plots may affect the results. The Arlington grove records were recomputed, shifting the control plots back one row. The yield of the first control plot (see Table XI) was 1,051 pounds instead of 800, and the last was 1,160 instead of 1,520 pounds. The first arrangement will be termed "arrangement A," the second "arrangement B." The mean yield of all control plots is not greatly changed by the different

arrangement. The mean of the "A" control plots is 1,367 pounds, that of the "B" control plots is 1,389 pounds, yet, as shown in Table XIV, the coefficient of variability is increased from 12.5 ± 1.1 in "arrangement A" to 18.2 ± 1.6 in "arrangement B" for single plots, with similar increases for the repeated plots.

TABLE XIV.—*Effect of two methods of arranging control plots*

Arrangement of plots of 10 trees.	Coefficient of variability.			
	Repeated once.	Repeated twice.	Repeated three times.	Repeated four times.
Arrangement A (Arlington)...	12.52 ± 1.07	7.53 ± 0.89	7.68 ± 1.17	4.33 ± 0.73
Arrangement B (Arlington)...	18.25 ± 1.59	16.07 ± 1.97	12.46 ± 1.91	8.75 ± 1.50

The records of Stockberger's hop yields have been computed in the same manner. The coefficient of variability of the plot yields in his arrangement is 16.71 ± 3.86. Moving the control plots down one—that is, using plot A as c_1 and so on—produces a coefficient of variability of 15.93 ± 3.55. There is obviously no difference between these two values.

The problem was also investigated by the use of the 8-tree plot lemon records. In this case three possibilities were tried, with the first, second, and third plots in turn as c_1 and every subsequent third plot as a control plot. The coefficients of variability for the different arrangements were 21.7 ± 2.21, 22.5 ± 2.29, and 23.8 ± 2.82. In view of the probable errors of the coefficients, there seems to be no real difference in the result of the different arrangements in this grove.

Since a decided difference was found in one case out of the three studied, it would seem that there is a rather high probability that significant differences may result from different arrangements of control plots.

VARIABILITY IN THE YIELDS OF MORE THAN ONE YEAR

It is often assumed that the mean yield of two or more years is less variable than the yield of one year.

We have had opportunity to study the variability in the yield of 60 navel-orange trees over a period of several years. The data were kindly furnished us by Mr. A. D. Shamel, of the Bureau of Plant Industry. The trees in question had been selected for their uniformly good production and the individual yields recorded for six years.

The figures presented in Table XV show that the variability of yields fell off distinctly after one year, but the reduction was negligible after the yields of two years were combined. It will be noted that the coefficient of variability of the single trees even for one year is notably low. Considering the records of single trees, the average of six years' records is not less variable than the average of two.

TABLE XV.—*Comparative variability of yields of navel-orange trees through a period of six years*

Class.	Coefficient of variability of the yield of—					
	1 year.	2 years.	3 years.	4 years.	5 years.	6 years.
Individual trees	22.8±1.5	15.0±0.9	14.7±0.9	13.3±0.8	13.7±0.9	14.6±1.0
Plots of 10 trees each	3.9±0.8	2.9±0.6	2.7±0.5	2.6±0.5	3.4±0.7	3.9±0.8

The coefficient of variability for the 10-tree plot is remarkably low for the reason that the trees themselves are so uniform and only a small area of ground is involved. It is no surprise, therefore, to find that the coefficients of variability are nearly equal, calculated from one to six years. The probable errors are relatively large and it is difficult to assert that there is any real difference.

The walnut yields can be used as additional data for the study of this question. Table XVI shows the coefficient of variability of the 1915 and 1916 yields and their total, with both 1- and 8-tree units. By considering the individual tree as a unit, the total yield for the two years was less variable than the 1916 yield, the difference equaling three times the probable error. The variability of the 1915 yield is practically equal to that of the total. On considering an 8-tree plot as a unit, there is a difference between the coefficients for 1915, 1916, and the total of the two years respectively; however, the observed difference is less than three times the probable error and its significance may be somewhat doubted. Apparently the mean of two years' yields in this case is less variable than one year's yield.

TABLE XVI.—*Comparative variability of yields of seedling walnut trees through a period of two years*

Class.	Coefficient of variability of the yield.		
	1915 yield.	1916 yield.	Mean of 1915 and 1916 yields.
Individual trees	47.9±1.6	53.9±1.9	46.4±1.6
Plots of eight trees each	30.0±2.6	33.3±3.0	25.3±2.2

Further studies on the comparison of the variability of yields through several years were made, from data published by Hedrick (*1911*). Table IV of the bulletin cited gives the yearly yields of individual apple trees from 1902 to 1910, inclusive, upon which our computations are based.

Four differential fertilizer treatments have been given to eight plots of five trees each, each treatment being duplicated on nonadjacent plots. There are three nontreated plots serving as controls.

The variability of the individual trees was computed on the 15 control trees.

The variability of plots is based on hypothetical plots made up of one tree from each of the treated and one from each of the untreated plots. These hypothetical plots are therefore made up of an equal number of trees having similar treatments. The variability of the 10 plots thus obtained was computed for the single year 1910, and for the sum of two, three, four, and seven years. Table XVII gives the coefficients of variability for single trees and for 5-tree plots.

TABLE XVII.—*Comparative variability of yields of Baldwin apple trees through a period of seven years* [a]

Class.	Coefficient of variability of the yield.				
	1 year (1910).	2 years (1909 and 1910).	3 years (1908–1910).	4 years (1906, 1908–1910).	7 years (1902, 1903, 1905, 1906, 1908–1910).
Individual trees..	37.3±5.2	33.6±4.6	32.5±4.4	32.2±4.4	34.0±4.7
Plots of five trees each.....	18.5±2.9	16.8±2.6	18.1±2.8	18.2±2.8	21.7±3.4

[a] Records taken from Hedrick (*1911, p. 172–174*).

It is interesting to note how slightly the variability of the yields is decreased by combining two or more years. If regard is paid to the probable errors, it can not be said that there is any real difference. In other words, one year's records of the yields of these apple trees seem to be as reliable for variation studies as those for several years.

It seems, therefore, that the continuation through several seasons may not so materially decrease the variability of tree yields as one might expect. This has a direct bearing on the reliability of the major portion of the calculations of this paper which are based on the variability of the yields of one season. It should be kept in mind, however, that these studies do not concern the relative yield of one plot compared with another, but rather deal with the total variation from the mean of the yields of all the plots. As cited before from the work of several experimenters, the relative productivity of a group of plots may not be fully determined even after a period of years, whereas the tree yields from Hedrick, Shamel, and the data of the writers indicate that a measure of the variability for one year of a group of trees divided into plots, may be very representative of the mean variability for several years.

SUMMARY

(1) The present paper is the result of a study of the nature and extent of the casual variability of yields of fruit trees under field conditions and its bearing on the reliability of plot trials.

(2) Studies have been made upon the variability of the yields of orange, lemon, apple, and walnut trees. The orchards studied were selected on

account of uniformity of treatment and appearance, yet the variability in productivity was considerable. The coefficient of variability for the yield of individual trees of the clonal varieties ranged from 29.27 ± 0.69 to 41.23 ± 1.52 per cent, but for the individual seedling walnuts, the coefficient was somewhat higher, reaching 53.91 ± 1.92 per cent. The variability of these tree yields approaches the normal curve of errors. This variability may be assumed to be the result of "casual" factors which are beyond the control and possibly the recognition of a careful experimenter.

(3) The effect upon variability of combining trees into plots of various sizes and shapes has been investigated. As the number of trees per plot is increased, the coefficient of variability decreases. The coefficient of variability does not decrease, however, in proportion to the increased number of trees per plot. In most cases there is little gained in accuracy by increasing the plot to include more than eight adjacent trees.

(4) One of the great causes of variability in yields appears to be the heterogeneity of apparently uniform soil. While a combination of a sufficient number of adjacent trees into a plot will overcome largely the fluctuations of individuals, nevertheless the plots may not sufficiently include both high- and low-yielding areas to give a typical average. Greater reliability may be secured by a systematic repetition and distribution of plots through the experimental area. A consistent gain in reliability resulting from this method of repetition is shown by the use of several different methods of computing the variability.

The coefficient of variability for an average plot of 16 adjacent trees was 22.58 ± 1.01, while 16 trees in four scattered ultimate plots each of four trees have a coefficient of variability of 9.29 ± 0.40. The larger the number of units in a combination plot the more typical is the sample of the area obtained. A 16-tree plot can be expected to give more reliable results if divided into four equal plots and repeated at four regularly placed intervals than can either two 8-tree plots, or 6 adjacent trees. The same principle holds true for larger units. A given number of unit plots will give a greater accuracy than half the number of units with twice as many trees per unit.

Four repetitions of an ultimate plot reduced the coefficient of variability to a point considered practical for cultural operations. Further repetitions, though reducing the coefficient in less degree, did not appear to justify the additional number of trees required. A minimum of 8 to 10 trees is required for plots involving cultural experiments. In the case of rootstock, pruning, or variety trials, twice as many plots each containing half as many trees might be used to obtain greater accuracy.

The fact that marked soil variations occur which tend to make adjacent trees or adjacent plots yield alike, even on soils which were chosen because of their apparent uniformity, is well shown by applying the formula proposed by Harris (*1915*) for measuring the coefficient of correlation between neighboring plots of the field. Applying this to the Arlington

navel oranges, the writers have calculated the correlation between the yield of the 8-tree plot as the ultimate unit and the yield of the combination of four such adjacent plots, and it was found that

$$r = +0.533 \pm 0.085.$$

This result shows a marked correlation, indicating a pronounced heterogeneity in the soil of this grove influencing fruit production.

However, when the correlation between the 8-tree plot as the ultimate unit and the yield of the combination of four such systematically scattered plots was calculated it was found that

$$r = +0.137 \pm 0.120.$$

This coefficient is practically equal to its probable error and can be regarded as significantly zero.

(5) In the computations made by the writers emphasis is also laid upon the nature and magnitude of the probable error. It is shown in several cases that the probable error of comparison between plots may be so large that relatively large differences must be evident between treated and untreated plots for a reasonable assurance that it is due to the factors being experimented upon. With the plots of 16 to 32 adjacent trees which were studied, a difference of from 62.94 to 81.97 per cent of the mean production would be necessary in order to obtain chances of 10 to 1 that the results were due to differential treatment and not to casual variation in the productivity of the trees. With the same number of trees in scattered units, a difference of 28.42 to 50.02 per cent would be necessary for the same odds. It seems probable, therefore, that a difference between two tree plots of less than 50 per cent of the mean production should be considered with caution before attributing it to differential treatment.

(6) The relation between the shape of a plot and its variability was investigated by making comparisons between square plots and linear plots containing the same number of trees. Except in the case of large plots, the difference in the variability of plots of different shapes was insignificant.

(7) In any method of field experimentation where a standard of comparison is desired the theoretical or "normal" yield of a plot is a question of importance. By the use of certain formulas the "normal" yield may be computed from control plots. As a standard, one may use the average yields of the control plots of the entire area, or of the nearest control plots, or a combination of the two. In cases studied, the coefficient of variability was reduced 50 per cent by calculating the normal yield from the nearest controls in place of using the mean of the entire area. The employment of every alternate row as a control plot was not sufficient to offset the variability due to soil heterogeneity.

(8) Computations made on the yields of orange, walnut, and apple trees for several consecutive years showed little annual fluctuation in their variability. One or two crops may not show greater variability than the average of six or seven crops.

LITERATURE CITED

BALLANTYNE, A. B.

1916. FRUIT TREE ROOT SYSTEMS. Utah Agr. Exp. Sta. Bul. 143, 15 p., 5 fig.

BARRE, H. W.

1915. APPLE BREEDING. *In* S. C. Agr. Exp. Sta. 28th Ann. Rpt. [1914]/15, p. 13–17.

CARLETON, M. A.

1909. LIMITATIONS IN FIELD EXPERIMENTS. *In* Proc. 30th Ann. Meeting Soc. Prom. Agr. Sci., p. 55–61.

COIT, J. E.

1910. THE RELATION OF ASEXUAL OR BUD-MUTATION TO THE DECADENCE OF CALIFORNIA CITRUS ORCHARDS. *In* Proc. 37th Fruit Growers' Conv. Cal., p. 32–39.

COOMBS, G. E., and GRANTHAM, J.

1916. FIELD EXPERIMENTS AND THE INTERPRETATION OF THEIR RESULTS. *In* Agr. Bul. Fed. Malay States, v. 4, no. 7, p. 206–216, 1 fig.

FLETCHER, S. W.

1913. ELIMINATING UNPROFITABLE TREES FROM THE APPLE ORCHARD. *In* Proc. 33d Bienn. Session Amer. Pomol. Soc., p. 138–145.

GOURLEY, J. H.

1915. STUDIES IN FRUIT BUD FORMATION. *In* N. H. Agr. Exp. Sta. Tech. Bul. 9, 79 p., 5 fig., 4 col. pl., 13 charts. Bibliography, p. 77–79.

HALL, A. D.

1909. THE EXPERIMENTAL ERROR IN FIELD TRIALS. *In* Jour. Bd. Agr. [London], v. 16, no. 5, p. 365–370.

——— and RUSSELL, E. J.

1911. FIELD TRIALS AND THEIR INTERPRETATION. *In* Jour. Bd. Agr. [London], Sup. 7, p. 5–14, 2 fig.

HARRIS, J. A.

1912. ON THE SIGNIFICANCE OF VARIETY TESTS. *In* Science, n. s., v. 36, no. 923, p. 318–320.

———

1913a. AN ILLUSTRATION OF THE INFLUENCE OF SUBSTRATUM HETEROGENEITY UPON EXPERIMENTAL RESULTS. *In* Science, n. s., v. 38, no. 975, p. 345–346, 1 fig.

———

1913b. SUPPLEMENTARY NOTE ON THE SIGNIFICANCE OF VARIETY TESTS. *In* Science, n. s., v. 37, no. 952, p. 493–494.

———

1915. ON A CRITERION OF SUBSTRATUM HOMOGENEITY (OR HETEROGENEITY) IN FIELD EXPERIMENTS. *In* Amer. Nat., v. 49, no. 583, p. 430–454.

HEDRICK, U. P.

1911. IS IT NECESSARY TO FERTILIZE AN APPLE ORCHARD? N. Y. State Agr. Exp. Sta. Bul. 339, p. 153–195, 7 fig., 4 pl.

———

1912. PEDIGREED NURSERY STOCK. N. Y. State Agr. Exp. Sta. Circ. 18, 8 p.

———

1914. A COMPARISON OF TILLAGE AND SOD MULCH IN AN APPLE ORCHARD. *In* N. Y. State Agr. Exp. Sta. Bul. 383, p. 249–281, 1 fig., 6 pl.

HOLTSMARK, G., and LARSEN, B. R.

1906. ÜBER DIE FEHLER, WELCHE BEI FELDVERSUCHEN DURCH DIE UNGLEICHARTIGKEIT DES BODENS BEDINGT WERDEN. *In* Landw. Vers. Stat., Bd. 65, Heft 1/2, p. 1–22.

KRAUS, E. J.

1916. SOMATIC SEGREGATION. *In* Jour. Hered., v. 7, no. 1, p. 3–8, 1 fig.

LEWIS, C. I, and VICKERS, H. A.

1915. ECONOMICS OF APPLE ORCHARDING. *In* Ore. Agr. Exp. Sta. Bul. 132, 104 p., 14 fig.

YON, T. L.

1912. SOME EXPERIMENTS TO ESTIMATE ERRORS IN FIELD PLOT TESTS. *In* Proc. Amer. Soc. Agron., v. 3, 1911, p. 89–114, 5 fig.

MACOUN, W. T.

1904. INDIVIDUALITY OF FRUITS. *In* Exp. Farms [Canada] Rpt., 1903, p. 102–104.

MERCER, W. B., and HALL, A. D.

1911. THE EXPERIMENTAL ERROR OF FIELD TRIALS. *In* Jour. Agr. Sci., v. 4, pt. 2, p. 107–132, 10 fig.

MITCHELL, H. H., and GRINDLEY, H. S.

1913. THE ELEMENT OF UNCERTAINTY IN THE INTERPRETATION OF FEEDING EXPERIMENTS. *In* Ill. Agr. Exp. Sta. Bul. 165, p. 459–579, 8 fig. Bibliography, p. 578–579.

MONTGOMERY, E. G.

1912. VARIATION IN YIELD AND METHOD OF ARRANGING PLATS TO SECURE COMPARATIVE RESULTS. *In* Nebr. Agr. Exp. Sta. 25th Ann. Rpt., [1911]/12, p. 164–180, 4 fig.

———

1913. EXPERIMENTS IN WHEAT BREEDING: EXPERIMENTAL ERROR IN THE NURSERY AND VARIATION IN NITROGEN AND YIELD. U. S. Dept. Agr. Bur. Plant Indus. Bul. 269, 61 p., 22 fig., 4 pl.

MUNSON, W. M.

1907. ORCHARD RENOVATION. *In* Maine Agr. Exp. Sta. Bul. 139, p. 53–56.

OLMSTEAD, L. B.

1914. SOME APPLICATIONS OF THE METHOD OF LEAST SQUARES TO AGRICULTURAL EXPERIMENTS. *In* Jour. Amer. Soc. Agron., v. 6, no. 4/5, p. 190–204, 1 fig.

PICKERING, S. U.

1911. EXPERIMENTAL ERROR IN HORTICULTURAL WORK. *In* Jour. Bd. Agr. [London], Sup. 7, p. 38–47.

PRITCHARD, F. J.

1916. THE USE OF CHECKS AND REPEATED PLANTINGS IN VARIETAL TESTS. *In* Jour. Amer. Soc. Agron., v. 8, no. 2, p. 65–81, 3 fig.

SHAMEL, A. D.

1912. BUD SELECTION AS A MEANS OF IMPROVING CITRUS AND OTHER FRUITS. *In* Ann. Rpt. Amer. Breeders' Assoc., v. 8, 1911, p. 497–503.

STEWART, J. P.

1913. FERTILIZATION AND CULTURE METHODS IN APPLE ORCHARDS. *In* Pa. Agr. Exp. Sta. Ann. Rpt. 1911/12, p. 497–563.

STOCKBERGER, W. W.

1916. RELATIVE PRECISION OF FORMULÆ FOR CALCULATING NORMAL PLOT YIELDS. *In* Jour. Amer. Soc. Agron., v. 8, no. 3, p. 167–175.

SURFACE, F. M., and PEARL, Raymond.

1916. A METHOD OF CORRECTING FOR SOIL HETEROGENEITY IN VARIETY TESTS. *In* Jour. Agr. Research, v. 5, no. 22, p. 1039–1050, 4 fig. Literature cited, p. 1050.

WHITTEN, J. C.

1915. BUD SELECTION FOR INCREASING YIELDS. *In* Mo. Agr. Exp. Sta. Bul. 131, p. 479–480.

WOOD, T. B.

1911. THE INTERPRETATION OF EXPERIMENTAL RESULTS. *In* Jour. Bd. Agr. [London], Sup. 7, p. 15–37, 2 fig.

——— and STRATTON, F. J. M.

1910. THE INTERPRETATION OF EXPERIMENTAL RESULTS. *In* Jour. Agr. Sci., v. 3, pt. 4, p. 417–440, 10 fig.

INTERRELATIONS OF FRUIT-FLY PARASITES IN HAWAII

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INTRODUCTION

The introduction and ultimate establishment of four species of larval parasites of the Mediterranean fruit fly (*Ceratitis capitata* Wied.) in Hawaii, wherein exist ideal conditions for the rapid and unchecked development of the host throughout the year, has opened an exceptional opportunity for an investigation of all phases of the work of these parasites, not only in their relations to the host but also to one another, and has made possible the determination of many points of unusual interest to biological students of insect parasitism and of particular value to entomologists dealing with considerations relating to general parasitic control of insect pests.[1]

The information herein presented is given, not as a final decision or positive argument against the introduction of many parasites of a single pest, but to reveal the actual need for careful biological studies of parasites, especially in their interactions upon one another, before general ntroductions or liberations can be intelligently undertaken.

Entomologists, detailed in the past for researches in insect parasitism in foreign countries, have customarily adopted the policy of assembling all available species of primary parasites of the insects under investigation, with the final intention of conveying them all to the home country for propagation and liberation. The chief caution of these workers has usually been the elimination of all secondary parasites from the material prepared for shipment. Admirable results have often been achieved. However, after the introduction of several species, through one or successive importations, few, if any, considerations have been given to the possibility of detrimental results arising from interference with the action of one parasite of primary importance and great prolificness by another of less value and proved inferiority.

The important bearing that a preliminary and detailed knowledge of parasite habits may have upon the general question of parasite importations has already been well directed to the attention of entomologists by Dr. L. O. Howard. He states[2] that—

> It is unwise and most unpromising to attempt heterogeneous and miscellaneous importations of parasites without careful study of the host insect on its home ground

[1] For a history of these parasitic introductions and a discussion of climatic and host relationships favoring parasitic increase, see BACK, E. A., and PEMBERTON, C. E., THE MEDITERRANEAN FRUIT FLY IN HAWAII. U. S. Dept. Agr. Bull. 536, 119 p., 21 pl., 24 fig. 1918.

[2] HOWARD, L. O. THE PRACTICAL USE OF THE INSECT ENEMIES OF INJURIOUS INSECTS. *In* U. S. Dept. Agr. Yearbook 1916, p. 282. 1917.

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and in its natural environment throughout the whole range of its existence and a similar biological study of its parasites and natural enemies under such conditions.

Some results of the recent work in Hawaii most strongly bear this out. Sufficient evidence has been obtained to throw serious doubt upon the assumption, often accepted, that the greater the number of species of parasites associated with the host, the greater the chances for its control. It is felt that the following data quite definitely indicate, at least in some cases, that better results may be obtained by a method of judicious selection of desirable species for introduction rather than by a wholesale and indiscriminate procedure. If 90 per cent of all individuals of an insect pest are destroyed by a single species of parasite, is it wise to attempt further control by bringing in other species, until it is known by positive and careful experimentation in the laboratory and field that these new species will not interfere with or check the normal activities of the first species?

In May, 1913, Prof. F. Silvestri succeeded in bringing two species of opiine parasites of the fruit fly into Hawaii. One, *Opius humilis* Silvestri, he brought from South Africa, and the other, *Diachasma tryoni* Cameron, was secured in Australia. Both were soon established in the Kona coffee district of the island of Hawaii. By 1915 it had become clearly evident that *O. humilis* was often parasitizing from 60 to 90 per cent of all of the fruit-fly larvæ developing in the coffee cherries. *D. tryoni* steadily but slowly increased and in time exhibited a capacity for occasionally parasitizing 50 per cent or more of the host larvæ. Here it is obvious that overlapping or duplication in parasitism was occurring. Clearly some fly larvæ were being stung by both species of parasites and frequently to a very considerable extent.

CANNIBALISM AMONG THE PARASITES

Early in 1916, Dr. E. A. Back, of the Bureau of Entomology, while examining the contents of some parasitized fruit-fly material from the field, observed under the microscope a larva of the parasite *Diachasma tryoni* attacking one of the parasite *Opius humilis*. A suspicion of disadvantageous consequences arising from complications attending the interactions of these parasites led Mr. C. L. Marlatt to assign to the writers an investigation of this subject.

Careful microscopical examinations of large numbers of fruit-fly larvæ and pupæ, collected from localities where both species of parasites were known to be well established and actively working together, soon revealed one striking fact. In the majority of cases where fruit-fly larvæ had been parasitized by both *Diachasma tryoni* and *Opius humilis* the latter was killed and the former developed to maturity. *O. humilis* is killed purely by wounds and lacerations inflicted upon it by the long, curved, sickle-like mandibles of the newly hatched larva of *D. tryoni*. This larva

attacks the young of *O. humilis*, usually at a point a few segments back of the head, the point of contact being clearly seen in Plate 12. Its mandibles open wide and snap into the body of the attacked larva spasmodically and with remarkable quickness. Often the entire operation of broadly opening and closing the mandibles may be almost imperceptible to the eye. It may move its entire body quickly, the caudal tip may be curled beneath the body and extended again suddenly, and the mandibles may be repeatedly opened and closed until successfully grasping the Opius larva. Besides possessing unusual powers for inflicting injury to other parasitic larvæ about it, it may avoid counterattack through ability to move quickly and through the protection afforded the entire ventral surface of the body by a thick mass of serosal, cellular material that accompanies the larva when it emerges from the egg, and which remains with it during its entire life in the first instar. This mass of cells may be seen clinging to Diachasma larvæ of the first instar in Plate 10.

The newly hatched larva of *Opius humilis* possesses mandibles which are also long and pointed, as shown in Plate 12, A, B. These may be used to good advantage when the larva is successful in bringing them in contact with individuals of its own or of other species of parasites. The larva, however, is sluggish, moves much less quickly than that of *Diachasma tryoni*, is protected ventrally by a much thinner, less adhesive mass of serosal cells, is much less capable of quick and powerful movement of the mandibles, and usually holds the body in a somewhat horizontal and exposed position. These deficiencies seem to explain its inability to avoid destruction by larvæ of *D. tryoni* or to offer successful counterattack when the two are lodged within the same host larva.

During the examination of nearly 3,000 fruit-fly larvæ or pupæ, parasitized in each case by the two species of opiines, the dead or dying and often struggling Opius larvæ were frequently dissected from the tissues of the host while still tightly clasped in the mandibles of the Diachasma larvæ. Plate 10, A, is reproduced from a photomicrograph of a larva of *O. humilis* actually within the grasp of an active, living larva of *D. tryoni*. In this particular case the operation of removing the two larvæ from the host, placing them upon a microscope slide in water, and covering them with a cover glass failed to separate them, and the Diachasma larva ultimately died with its mandibles deeply embedded in the body of the dead Opius larva in the exact position as shown. Plates 12 and 13, A, B, C, show larvæ of *O. humilis* in various stages of laceration and distortion just as they were removed from fruit-fly larvæ or pupæ in which were also one or more larvæ of *D. tryoni*.

Extensive laboratory experiments have exactly confirmed the results of the first series of field observations. The aggressive, cannibalistic period of activity of the larvæ of these parasites is during existence in

the first instar, and particularly during the early period of this stage, before the body becomes engorged and swollen with food. Plate 11, B, which shows a mature first-instar larva of the parasite *Diachasma tryoni*, is interesting in this connection when compared with the other illustrations of newly hatched Diachasma larvæ of this instar. The head is unchanged in size, but the body is greatly distended after two days of feeding preparatory to molting to the second instar. In all of the illustrations the enlargement is the same. It is obvious that the enlarged and somewhat rigid body of the well-fed larva permits much less freedom of movement than is possible shortly after hatching. The body is first elastic, flexible, and capable of quick and effective action. This change in the size of the body may occur in from two to two and one-half days.

The molt to the second instar still further incapacitates the opiine larva for carrying on further cannibalistic action. The mandibles are then small, soft, and are almost imperceptible, even under high magnification, because of their transparency, being wholly unfitted for active use except in the separation of the semiliquid media in which the larva lies after the host has formed the puparium. The strong and heavily chitinized head of the opiine larva in the first instar is thus entirely discarded upon the molt to the second instar, and all further cannibalism ceases. The helpless condition after the first molt is well suggested in Plate 11, A, which shows a larva of *Diachasma tryoni* in the second instar.

During 1916 a microscopical examination of the contents of a total of 2,925 parasitized fruit-fly larvæ and pupæ definitely showed that the parasites readily oviposit in the same host larva more than once and exhibit no discernible instinct of selection of parasitized or unparasitized larvæ. All eggs so deposited hatch. Thus, fly larvæ commonly opened were found to contain several eggs or larvæ of the two species of parasites and sometimes a third species (*Diachasma fullawayi*), a later introduction.

Here was certain evidence of a complicated overlapping or duplication of parasitism. In no single instance were two parasite larvæ ever observed to develop to maturity in the same host larva, except in the case of the chalcid *Tetrastichus giffardianus* (p. 292). All but one were killed while still in the first instar, or occasionally before hatching. At times from 8 to 10 opiine larvæ were found within the host larva. Only one would mature and, as a rule, if a larva of Diachasma were one of the number, it survived all others.

If several eggs of *Diachasma tryoni* or *D. fullawayi* alone are deposited in an individual host larva over a period of two or three days, the last parasite larva to hatch stands the best chance for destroying the others and maturing. The case is identical with the *Opius humilis*. However, a well-grown and fully-fed larva of *D. tryoni* or *D. fullawayi*, if still in the

first instar, seems entirely able to destroy larvæ of *O. humilis* that are newly hatched and unencumbered with a body engorged with food.

All opiine larvæ within an individual fruit-fly larva or pupa may sometimes be fatally wounded and no parasite develop. This is not frequent. Often the deposition of 8 or 10 parasite eggs into a single fly larva causes its death. In such cases the parasite eggs usually hatch and the resulting larvæ die within a short time.

Cases occurred in which as many as 10 dead larvæ of *Opius humilis* were dissected from a fruit-fly larva, together with a single, vigorous, active larva of *Diachasma tryoni*. This does not necessarily mean that all of the 10 larvæ were killed by the latter. No doubt some of the former larvæ destroyed each other, but it clearly shows the superior aggressive and defensive power of the larva of *D. tryoni*. Many cases have been observed in which a larva of *O. humilis* was badly cut and distorted from attack by a larva of *D. tryoni*. Some such cases are shown in Plates 10, 12, and 13. Occasionally the body may be found entirely severed from the head. These extreme cases are no doubt caused by reattack upon the larva a day or more after it has died and has become somewhat softened. In most cases the death of the larva seems to be caused by the first grasp or pinch of the attacking larva. A single perforation in the body wall should be sufficient to cause the death of the larva in a short time.

Occasionally an opiine larva will destroy eggs of its own kind when it occurs in the same individual host with the eggs. In this manner mature embryos are sometimes very much distorted and almost unrecognizable. This is not frequently seen.

Cool weather materially retards the development of the opiine egg, particularly in the case of *Diachasma tryoni* and *D. fullawayi*. At such times many cases have been observed in which larvæ of *Opius humilis* have developed to the second instar before an egg of a species of Diachasma, which had been deposited into the same host larva harboring *O. humilis*, had hatched. Upon hatching the small, active Diachasma larva quickly destroyed the large, bulky Opius larva. This unusual condition has been observed only in January in the cool, elevated, coffee districts on the Island of Hawaii.

From over 2,900 cases where parasitized fruit-fly puparia have been opened, in no single instance has a case been observed in which the host pupa was formed. A host larva once parasitized quite readily forms into a normal puparium when sufficiently developed, but the presence of a single small opiine egg within its body invariably prevents any further development. The puparium is formed, the histolysis of the tissues is completed, and here all development of the fruit fly ceases. The broken-down and liquid medium thus prepared within the puparium, in which the parasite larvæ may move about and feed, enables them to reach all portions within and easily to come in contact with any other parasitic individuals that may occur there with them.

Thus, the inevitable tendency of every individual opiine larva upon hatching is to destroy every other parasitic larva in its domain, whether it be one of its own kind or of another species. This would appear to be an infallible instinct and one of great consequence in Hawaii in the development of the opiine parasites now present.

SUPPRESSION OF OPIUS HUMILIS BY DIACHASMA TRYONI AND D. FULLAWAYI

The parasites *Diachasma tryoni* and the closely related *Diachasma fullawayi*, by virtue of this larval instinct, coupled with further endowed superior body characters, have been responsible for the great suppression of the parasite *Opius humilis*. The last-named species is more prolific and hardy than either of the two others and is more generally efficient than both combined. By their association with the *O. humilis*, they have worked a detriment by reducing the total extent of parasitism to a point below that to which it is capable of exerting alone. The evidence of such suppression, gained from microscopical examinations of fruit-fly larvæ and pupæ secured from various fruits in Hawaii during 1916 and 1917, may be expressed as follows:

From April 16 to May 10, 1916, a dissection was made of 757 fruit-fly pupæ, freshly secured from coffee collected in the Kona coffee district of the Island of Hawaii. From this total, 345 were parasitized by only *Opius humilis*, 90 contained living larvæ of *Diachasma tryoni* together with dead larvæ of *O. humilis*, 9 contained living larvæ of *O. humilis* together with dead larvæ of *D. tryoni*, 1 was parasitized by only *D. fullawayi*, 5 contained living larvæ of *D. fullawayi* together with dead larvæ of *O. humilis*, 2 contained living Opius larvæ together with dead larvæ of *D. fullawayi*, 57 contained living larvæ of only *D. tryoni*, and 248 were not parasitized. Here it is seen that from the total of 757 puparia, 106 cases of duplication in parasitism occurred in which a species of Diachasma was found in the same puparium with one or more individual larvæ of *O. humilis*, and that in 95 of these the latter was killed and the former survived.

A further series of microscopical examinations of the contents of fruit-fly puparia, freshly secured from coffee fruits in this same district in January, 1917, during the coolest part of the year, strongly confirmed the results of the previous year. Six hundred and twenty-seven puparia were opened and examined. Of these a total of 343 were parasitized by *Opius humilis* alone, 67 were parasitized by only *Diachasma tryoni*, 2 contained only larvæ of *Diachasma fullawayi*, 129 contained living larvæ of *D. tryoni* together with dead larvæ of *O. humilis*, 8 contained dead larvæ of *D. tryoni* together with living larvæ of *O. humilis*, 4 contained living larvæ of *D. fullawayi* together with dead larvæ of *O. humilis*, 2 contained dead larvæ of *D. fullawayi* together with living larvæ of *O. humilis*, and 72 puparia were unparasitized. These results are again very significant.

From the total of 627 puparia 143 cases are noted in which an overlapping in parasitism occurred, wherein the puparia in each case contained larvæ of *O. humilis* in combination with larvæ of a species of Diachasma, and in 133 of these cases the former was destroyed.

The collection of extensive data on the percentage of parasitism exerted by the fruit-fly parasites in Hawaii over a period of three years gave abundant proof that the parasite *Diachasma tryoni* was most active during the warmer months of the year. This increase in activity and abundance paralleled a reciprocal decrease in the abundance of *Opius humilis*. The reverse was true during the remainder of the year, when the former species rapidly decreased and the latter ascended to a place of first importance. The data presented in Table I most positively reveal the extent of fluctuation in the comparative abundance of *O. humilis* and *D. tryoni*, the effectiveness of *O. humilis* being clearly at its maximum during the spring, when the abundance of *D. tryoni* is at its lowest, owing to the accumulated effect of the cool winter months.

TABLE I.—*Comparison of seasonal abundance of Opius humilis and Diachasma tryoni*

Locality.	Date of collection of host.[a]	Number of *Diachasma tryoni* emerging.	Number of *Opius humilis* emerging.	Percentage of *Diachasma tryoni.*	Percentage of *Opius humilis.*
Honolulu, Oahu	Mar., 1916	30	1,000	2.9	97.1
Do	Apr., 1916	1,200	9,778	10.9	89.1
Do	May, 1916	499	2,127	19.0	81.0
Do	June, 1916	2,303	998	69.8	30.2
Do	July, 1916	1,786	549	76.5	23.5
Do	Aug., 1916	2,286	649	77.9	22.1
Do	Sept., 1916	4,514	1,139	79.9	20.1
Do	Oct., 1916	6,772	2,061	76.7	23.3
Do	Nov., 1916	4,451	1,206	78.9	21.1
Do	Dec., 1916	2,605	1,602	61.9	38.1
Do	Jan., 1917	1,406	679	67.4	32.6
Do	Feb., 1917	558	1,101	33.6	66.4
Kona District, Hawaii	Sept., 1915	330	118	73.6	26.4
Do	Dec., 1915	210	274	43.3	56.7
Do	Mar., 1916	85	440	16.1	83.9
Do	Apr., 1916	756	3,031	19.9	80.1
Do	Aug., 1916	271	77	77.9	22.1
Do	Jan., 1917	558	4,749	10.5	89.5

[a] For similar data for 1915, see BACK, E. A., and PEMBERTON, C. E., THE MEDITERRANEAN FRUIT FLY IN HAWAII. U. S. Dept. Agr. Bul. 536, 119 p., 21 pl., 24 fig. 1918.

From September to December, 1916, a microscopical examination was made of the contents of 618 fruit-fly puparia obtained about Honolulu from coffee, guavas, and the winged kamani (*Terminalia catappa*). Of this total, 55 puparia were parasitized by only *Opius humilis*, 331 were parasitized by only *Diachasma tryoni*, 35 contained only larvæ of *D. fullawayi*, 96 contained living larvæ of *D. tryoni* together with dead larvæ of *O. humilis*, 4 contained dead larvæ of *D. tryoni* together with living larvæ

of *O. humilis*, 6 contained living larvæ of *D. fullawayi* together with dead larvæ of *O. humilis*, 1 contained a dead larva of *D. fullawayi* together with a living larva of *O. humilis*, and 90 were unparasitized. Here again is striking evidence of positive suppression of the parasite *O. humilis* by the other parasites, particularly by *D. tryoni*. Of the 618 puparia examined, the *O. humilis* occurred 107 times in combination with a species of Diachasma and won the struggle for existence in only 5 cases.

The foregoing would tend to explain the very noticeable fluctuations in the comparative abundance of the two principal species of parasites at differenct seasons of the year, as shown in Table I. It is seen that the extent of parasitism by *Opius humilis* is greatly influenced by the abundance or scarcity of the parasite *Diachasma tryoni*, and, as elsewhere discussed, the abundance of this latter parasite in Hawaii is very much dependent upon seasonal conditions. This causes a seasonal rise and fall in the effectiveness of *O. humilis*. In the summer and autumn of the year the ascendancy of the *D. tryoni* causes a great reduction in the abundance of *O. humilis*. During the winter and spring seasons the reduced activity of the Diachasma permits a rapid increase in parasitism by *O. humilis*. This is particularly true in the elevated Kona coffee district, where the winter temperatures are somewhat below those about Honolulu.

The results of the first series of dissections of fruit-fly larvæ and puparia on the island of Hawaii in January, 1916, led to further laboratory experiments in Honolulu on a comprehensive scale, duplicating as closely as possible the field conditions. Unparasitized fruit-fly larvæ were exposed within fruit to the attack of both *Opius humilis* and one or both of the species of Diachasma for a few hours. The fruit-fly puparia thus obtained were usually all parasitized, and no loss in time resulted from examinations of unparasitized material. In this manner 393 cases were obtained in which fruit-fly larvæ were parasitized by both *O. humilis* and *D. tryoni*. In 387 of these the *Opius* larvæ were all killed and the Diachasma survived. In only 5 of the puparia did the former succeed in overcoming the latter and developing.

Out of 77 cases in which fruit-fly larvæ were parasitized by both *Opius humilis* and *Diachasma fullawayi*, in only 2 did the Opius develop. In the remaining 75 the Opius larvæ were all killed by the Diachasma larvæ.

SUPPRESSION OF OPIUS HUMILIS BY TETRASTICHUS GIFFARDIANUS

The fruit-fly parasite *Tetrastichus giffardianus* Silvestri, a late introduction into Hawaii, has proved decidedly destructive to any of the opiines when occurring in the same fly larvæ or puparia with them. This chalcid, seemingly of importance, has, after a two years' trial in Hawaii, given but small promise of accomplishing any perceptible control of the

fruit fly. It is occasionally bred out from fruit-fly material secured in the field. Several times during dissections of puparia from the field its larvæ have been found in combination with larvæ of *Opius humilis* or of one of the species of Diachasma. Though soft, sluggish, and armed with small, inconspicuous, blunt mandibles, it nevertheless survives the opiine larvæ by sheer force of numbers and consequent rapid absorption of food. When ovipositing, this chalcid usually places about 10 eggs in the host at one insertion of the ovipositor. The opiines deposit but one egg, remove the ovipositor, and look for further larvæ to attack. A single oviposition, then, by a *T. giffardianus* into a host larva already parasitized by an opiine, places about 10 individuals of the chalcid with 1 of the opiine. If the host larva has already received several opiine ovipositions, usually only one individual is alive, as already shown. The larvæ of *T. giffardianus* exhibit no cannibalistic tendencies, and do not destroy each other. Many of the chalcid larvæ are then killed by the opiine larvæ, but seldom all of them. The opiines ultimately die, and one or more of the Tetrastichus larvæ develop. The death of the Opius or Diachasma larvæ results usually from starvation or suffocation or possibly by the absorption of toxic excretions of the Tetrastichus larvæ. Certainly the chalcid larvæ inflict no visible bodily injury on the opiine larvæ.

In view of the demonstrated ineffectiveness of this chalcid and of its capability for surviving the *Opius humilis* when occurring in fruit-fly larvæ with it, it is here considered a detrimental introduction because of interference with the work of the latter.

OPIUS HUMILIS, THE MOST PROLIFIC OF THE INTRODUCED PARASITES

From the foregoing summaries of the data it is obvious that the parasite *Opius humilis* is killed in the larva stage in almost every instance in which its larvæ become associated in a host larva with any one of the three other species of introduced fruit-fly parasites and that the percentage of cases of such duplication is large. Biological studies of all of these parasites, so far as two and three years' adaptation in a new country may show, have indicated quite clearly that under Hawaiian conditions *O. humilis* is the most prolific of the four species in all seasons of the year and that none of the others show particular abilities for reaching larvæ of the fruit fly that are not as easily accessible to the attack of *O. humilis*.

The very considerable activity of the parasite *Diachasma tryoni* and the occasional heavy parasitism by it has made necessary the establishment of careful proof that it is less prolific than *Opius humilis*, before it can be maintained that the introduction of the former is a detrimental one. The unimportance of the other two parasitic species eliminates all present need for discussing them any further.

From what has already been shown, we know positively that the parasite *Diachasma tryoni* has a clear field in Hawaii for unchecked

reproduction. *Opius humilis*, though present everywhere, does not hinder its activities. Extensive records kept in Hawaii during a period of three years on emergences from more than 100,000 fruit-fly puparia from many sections of the islands and from all available types of fruits have clearly shown that the average maximum degree of parasitism by *D. tryoni* does not exceed or equal that of *O. humilis*. This alone is sufficient proof of superior prolificness on the part of the latter.

In no instance have fruits of all varieties been found harboring larvæ of the fruit fly that are less frequented by *Opius humilis* than by *Diachasma tryoni*. The ability of the former to find the host is, then, equal, if not superior, to that of the other parasite. In no case has evidence been found to show that the longer ovipositor of the *D. tryoni* is really an advantage over that of the *O. humilis*, whose ovipositor is less than a third as long. The comparative difference is considerable, but the difference when considered in fractions of an inch is really small.

Preliminary records of individual females of both species to determine the total number of eggs deposited by an individual in a lifetime show no superiority of the *Diachasma tryoni* over *Opius humilis* in this respect.

The life cycle of *Diachasma tryoni* is consistently longer than that of *Opius humilis*. From 28,410 records on the length of the life cycle of the former, secured during 1916 and 1917, it is almost invariably found to be from two to four days longer than that of the latter during most of the year, and in the winter months a great number of the Diachasma individuals, hibernating in the larva stage, extended the cycle to from one to six months longer. From 22,889 cases under observation in 1916 and 1917 on the life cycle of *O. humilis*, in no single case has an individual ever been known to so hibernate or extend the length of the life cycle beyond the average for more than a few days. Cool weather and drouth seem most favorable for inducing this hibernating tendency in the larvæ of the *D. tryoni*. Of 3,077 cases under observation by the junior author in January, February, and March, 1917, in which fruit-fly larvæ had been parasitized by a Diachasma, a total of 1,404 cases occurred in which the parasite went into hibernation as a mature larva. This seems to explain the great reduction in abundance of *D. tryoni* in the field in winter. Its capacity as a parasite in the winter months is thus strikingly less than that of *O. humilis*.[1]

From the standpoint of longevity, all experiments so far show no great superiority of one species over the other, except as noted above in regard to hibernation. Individual adults of both *Opius humilis* and *Diachasma tryoni* have been kept alive for about four months.

[1] Credit is here due Mr. J. C. Bridwell for valuable suggestions offered in connection with the study of the hibernation of these parasites.

The relative proportion of females to males in these two species of parasites is interesting. Of 26,975 individuals of *Diachasma tryoni*, reared from material collected in the field in 1916 and 1917, 16,845, or 62.4 per cent, were males. Of 10,843 individuals of *Opius humilis* similarly reared from material collected in the field in 1916 and 1917, 6,128, or 56.5 per cent, were males. Here, again, the advantage, if any, lies with *O. humilis*.

In view of these several facts relating to the comparative prolificness of the parasites *Opius humilis* and *Diachasma tryoni*, it appears that the former is superior to the latter species or to any of the other introduced parasites. Thus, when a host larva is parasitized by both *O. humilis* and *D. tryoni*, the latter survives, and in so doing produces an individual less prolific than would have been the case had the *O. humilis* been permitted to develop. This seems to point to a certain, distinct loss. If one species of larval parasite when working alone parasitizes 60 per cent of the host, and another species not strikingly different from the first and working the same in every known respect parasitizes 40 per cent of the host when working alone, there is no reason to assume that both combined can exceed a parasitism of 60 per cent. All overlapping by the species capable of only 40 per cent parasitism can only serve to reduce the total effect to a point below 60 per cent of parasitism.

CONCLUSION

Sufficient evidence has been presented to prove the superiority of the parasite *Opius humilis* over the other introduced fruit-fly parasites in Hawaii and demonstrates the decided restraint operated over it by the unfailing cannibalistic activities of the larvæ of *Diachasma tryoni* in particular and of the other parasites in part. Knowing the capacity of *O. humilis* for parasitizing from 80 to 90 per cent of the larvæ of the fruit fly in favorable localities, such as the large Kona coffee belt on the island of Hawaii, the writers here maintain that detrimental results to a certain extent have arisen from the liberation in Hawaii of parasites other than *O. humilis* that attack the larva of the fruit fly. The total parasitism has simply been reduced in value to that of a parasite of secondary value.

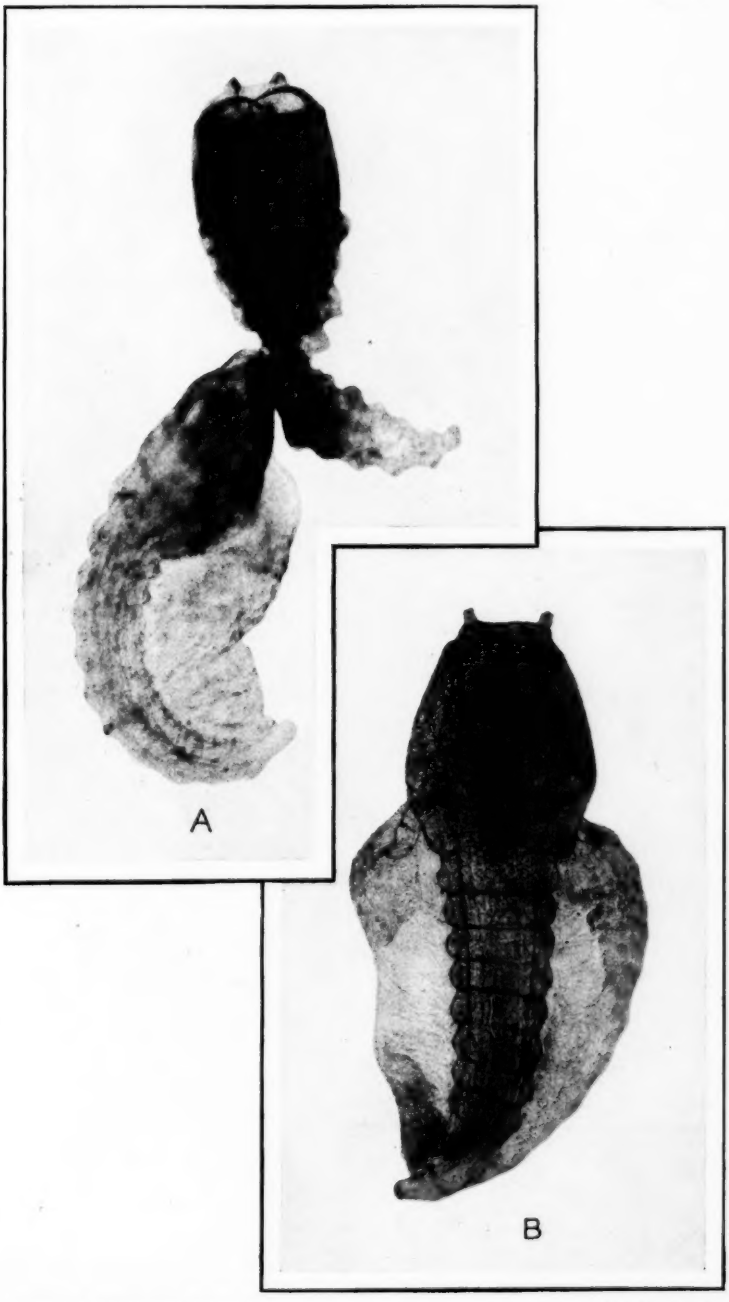
It is hoped that the present analysis of the interrelated activities of the imported fruit-fly parasites in Hawaii may serve to stimulate greater discrimination in the selection of parasites proposed for future introduction.

PLATE 10

Diachasma tryoni:

A.—Freshly hatched larva with its mandibles actually embedded in the body of a newly hatched but dead larva of *Opius humilis*. ×100.

B.—Newly hatched larva with its mandibles closed, showing ventral serosal material surrounding the body and the two gill-like appendages on the first body segment. × 100.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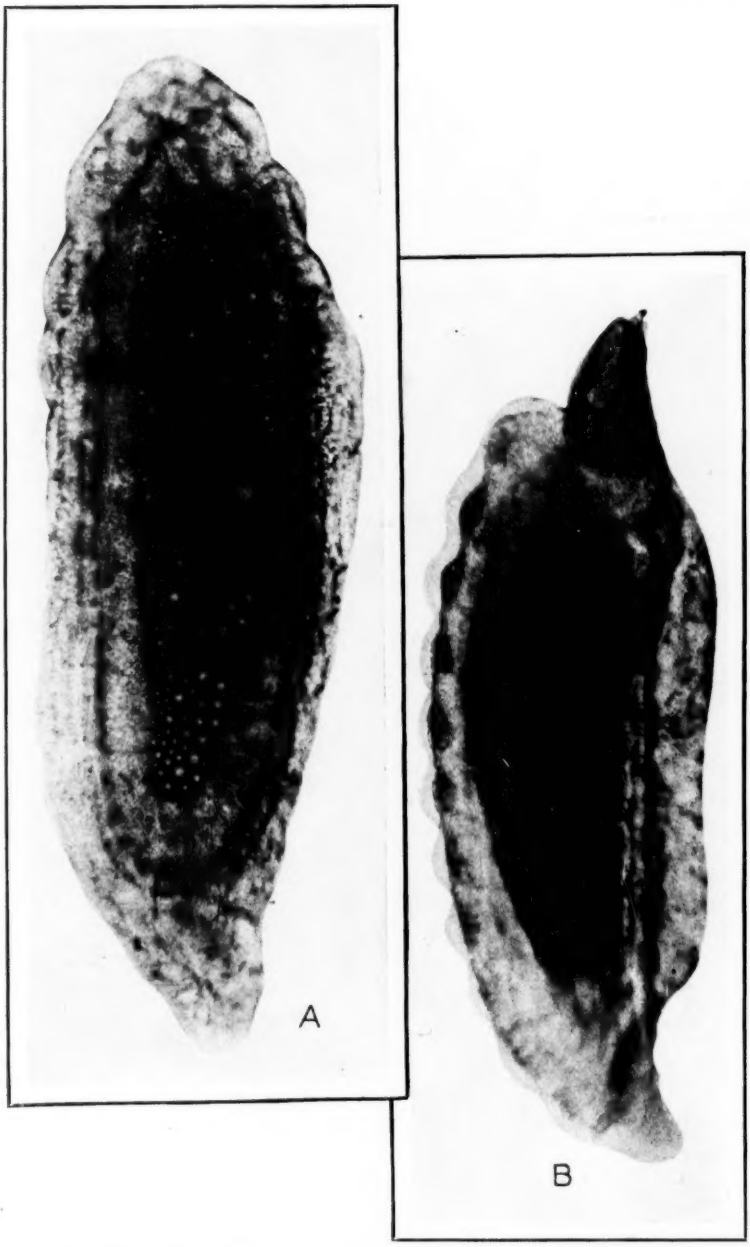
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PLATE 11

Diachasma tryoni:

A.—Lateral view of larva in the second instar, showing particularly well the fat-body of the host recently taken in as food. × 100.

B.—Lateral view of a 2-day-old larva engorged with food and about to molt, showing the enlarged and stiffened body. × 100.

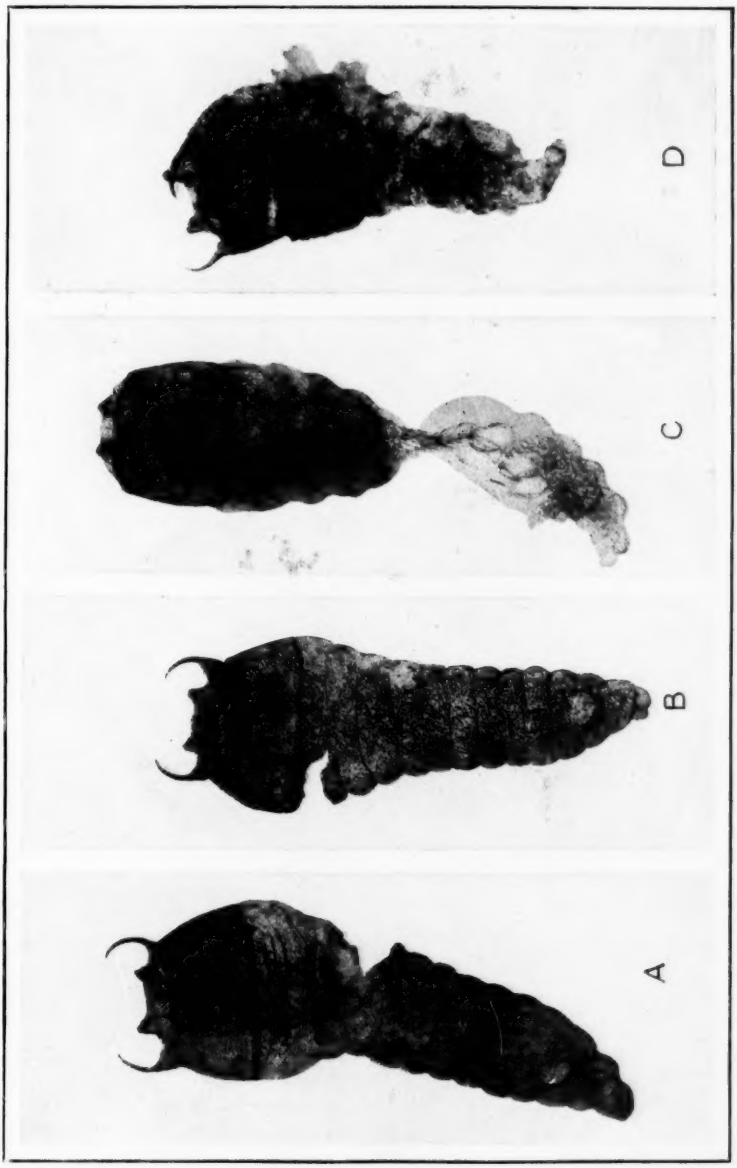
PLATE 12

Opius humilis:

A, B.—Dead larva in first instar; killed by first-stage larva of *Diachasma tryoni*, showing cut on body made by the attacking larva and mandibles extended in final death struggle. × 100.

C.—Dead larva in first instar; killed by first-stage larva of *Diachasma tryoni*. The point of attack is here clearly seen. The body contents have been apparently withdrawn from the lower portion of the body by the attacking larva. × 100.

D.—Dead larva in first instar; badly lacerated and distorted by attack of first-stage larva of *Diachasma tryoni*. In this case the dead larva was probably destroyed while n the embryonic stage and a few hours prior to the hatching of the egg. × 100.


D
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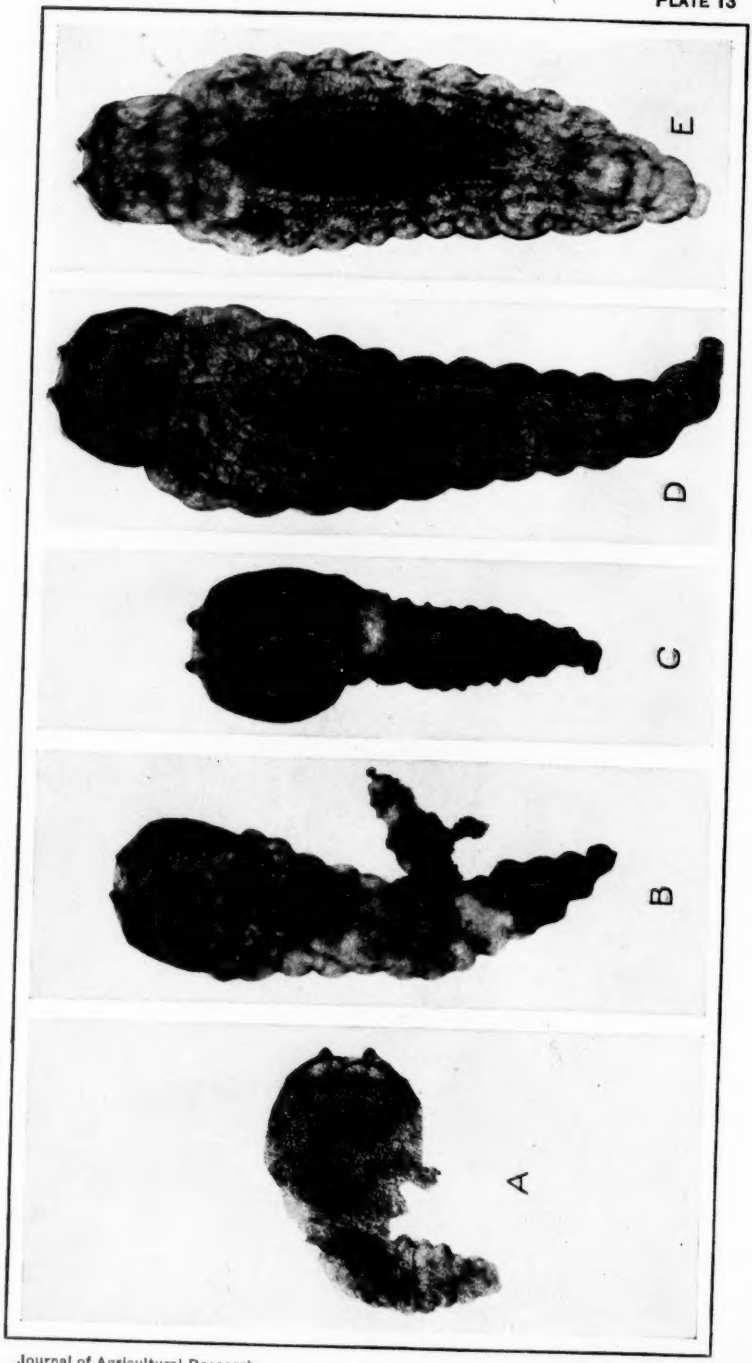
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PLATE 13

Opius humilis:

A.—Dead larva in first instar, with body shriveled and twisted through attack by first-instar larva of *Diachasma tryoni*. × 100.

B.—Dead larva in first instar; killed by first-instar larva of *Diachasma tryoni*. Here the larva had been feeding and developing for several hours before being attacked by the Diachasma larva. The body contents can be seen protruding from an inflicted wound on the seventh and eighth body segments. × 100.

C.—Dead larva in first instar; killed by first-stage larva of *Diachasma tryoni*. This is the appearance of the dead Opius larvæ most commonly seen. The body is pinched by the mandibles of the Diachasma larva in the first or second body segment back of the head. × 100.

D.—Healthy, living larva in first instar. × 100.

E.—Healthy, uninjured, living larva in first instar. × 100.